



The 18th EURL-AR Proficiency Test - Enterococci, Staphylococci and E. coli 2015

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Publication date:
2016

Document Version
Publisher's PDF, also known as Version of record

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Citation (APA):
Cavaco, L., Karlsmose Pedersen, S., Hendriksen, R. S., & Aarestrup, F. M. (2016). *The 18th EURL-AR Proficiency Test - Enterococci, Staphylococci and E. coli 2015*. National Food Institute, Technical University of Denmark. http://eurl-ar.eu/data/images/reports/eurl%20report_eqas_ec_st_ent_isbn_inclapp.pdf

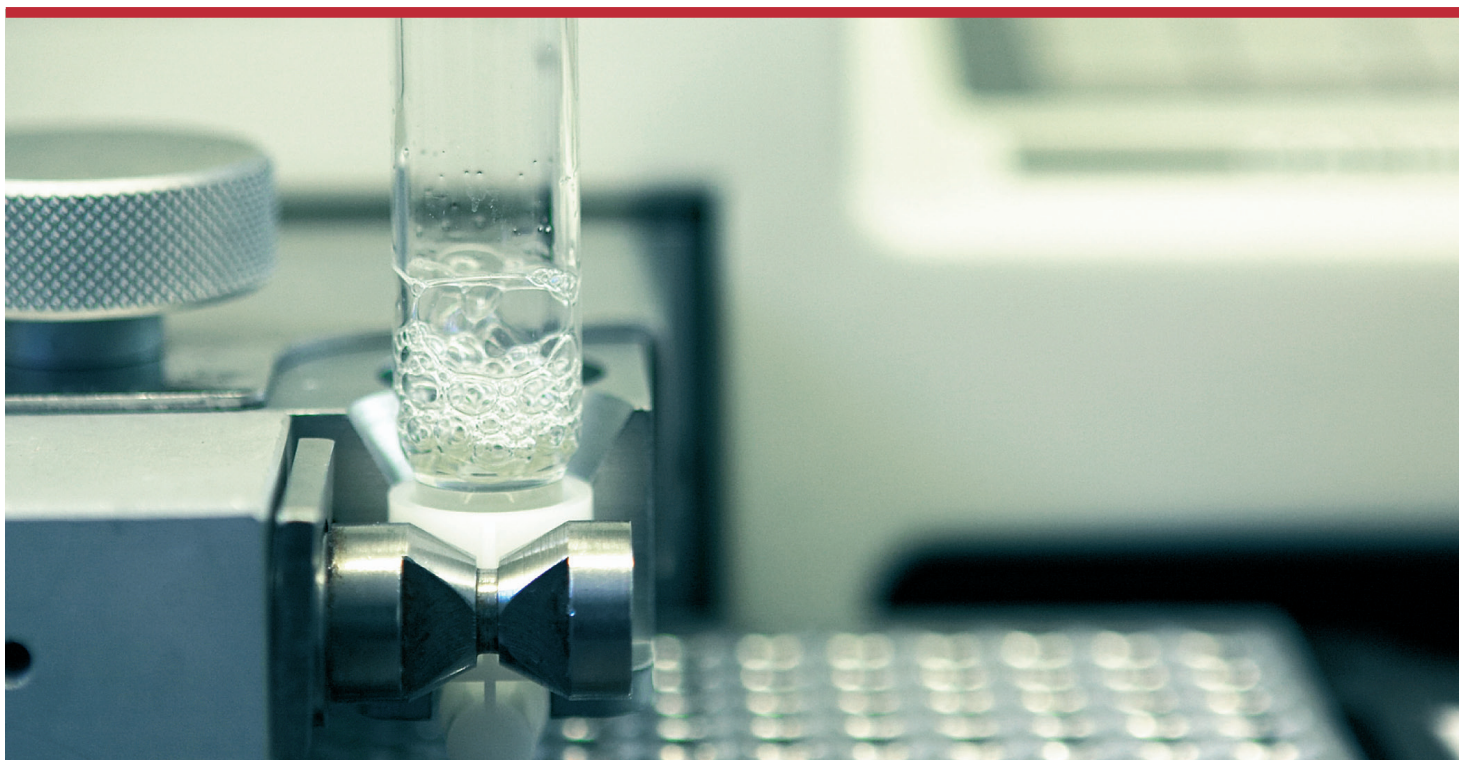
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The 18th EURL-AR Proficiency Test - Enterococci, Staphylococci and *E. coli* 2015



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DTU Food
National Food Institute



**THE 18TH EURL-AR Proficiency Test Enterococci, Staphylococci and
Escherichia coli - 2015**

1. edition, June 2016

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ISBN: 978-87-93109-85-8

The report is available at

www.food.dtu.dk

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1. Introduction

This report is describing the results of the eighteenth proficiency test organized by the National Food Institute as the EU Reference Laboratory for Antimicrobial Resistance (EURL-AR). This proficiency test focuses on antimicrobial susceptibility testing (AST) of enterococci, staphylococci and *Escherichia coli*. It is the ninth External Quality System Assurance System (EQAS) conducted for these microorganisms.

The aim of this EQAS is to: i) monitor the quality of AST results produced by National Reference Laboratories (NRL-AR), ii) identify laboratories which may need assistance to improve their performance in AST, and iii) determine possible topics for further research or elaboration.

When reading this report, please take into account the following important considerations:

1) Expected results were generated by performing Minimum Inhibitory Concentration (MIC) determinations for test strains in two different occasions at the Technical University of Denmark, National Food Institute (DTU-FOOD). These results were then verified by the United States Food and Drug Administration (FDA), Centre for Veterinary Medicine. Finally, a fourth MIC determination was performed at DTU-FOOD after preparation of the agar stab culture for shipment to participants to confirm that the vials contained the correct strains with the expected MIC values.

2) The evaluations are based on interpretations of AST values determined by the participants. This is in agreement with the methods included in the EU Decision 652/2013 which are to be used for the testing of *E. coli* and Enterococci species and the most recent recommendations from EFSA regarding the testing of *Staphylococcus aureus* by AST. The methods used by the participants should be reflecting

those used to report AST data to the European Food Safety Authority (EFSA), so that they comply with “the main objective of this EQAS; to assess and improve the comparability of surveillance and antimicrobial susceptibility data reported to EFSA by the different NRLs”, as stated in the protocol.

3) Given the EU regulation referring only to the use of MIC methods, and the set-up of the database, the reporting of Disk diffusion data was not allowed.

4) The EURL-AR network has previously agreed on setting the accepted deviation level for laboratory performance to 5%.

Evaluation of a result as “deviating from the expected interpretation” should be carefully analyzed in a self-evaluation procedure performed by the participant right after the EQAS trial results are disclosed. Since methods used for MIC determination have limitations, it is not considered a problem to obtain a one-fold dilution difference in the MIC of a specific antimicrobial when testing the same strains. However, if the expected MIC is close to the breakpoint value for categorizing the strain as susceptible or resistant, a one-fold dilution difference, which is acceptable, may result in two different interpretations, i.e. the same strain will be categorized as susceptible and resistant, which will be evaluated as correct in one case and incorrect in the other if the evaluation is based on interpretation of MIC values. Since this report evaluates the interpretations of AST values, some participants may find their results classified as wrong even though the actual MIC they reported is only one-fold dilution apart from the expected MIC. In these cases, the participants should be confident about the good quality of their performance of AST. In the organization of the EQAS we try to avoid these situations by choosing test strains with MIC

values more distant from the breakpoints for resistance, which is not always feasible for all strains and antimicrobial combinations. For this reasons, the EURL-AR network unanimously has established in 2008 that if there are less than 75% correct results for a specific strain/antimicrobial combination, the reasons for this situation must be further examined and, on selected occasions explained in details case by case, these results may subsequently be subtracted from the evaluation report.

This report is approved in its final version by a technical advisory group composed by competent representatives from all NRLs who meet once a year at the EURL-AR workshop.

All conclusions presented in this report are publically available. However, participating laboratories are identified by codes and each code is known only by the corresponding laboratory. The full list of laboratory codes is confidential information known only by relevant representatives of the EURL-AR and the EU Commission.

The EURL-AR is accredited by DANAK as provider of proficiency testing (accreditation no. 516); working with zoonotic pathogens and indicator organisms as bacterial isolates (identification, serotyping and antimicrobial susceptibility testing).

2. Materials and Methods

2.1 Participants in EQAS 2015

A pre-notification to announce the EQAS 2015 on AST of enterococci, staphylococci and *E. coli* was sent by e-mail on the 5th May 2015 to the designated NRLs in the network (App. 1) and including eight additional laboratories (Denmark, Iceland, Norway, Serbia, Spain,

Switzerland, The Netherlands and Turkey). These were invited to take part in the EQAS 2015 on the basis of their participation in previous EQAS iterations and/or affiliation to the EU network. Finally, the participant laboratories in the EQAS represented all EU Member States and three laboratories represented non-MS Norway, Switzerland and

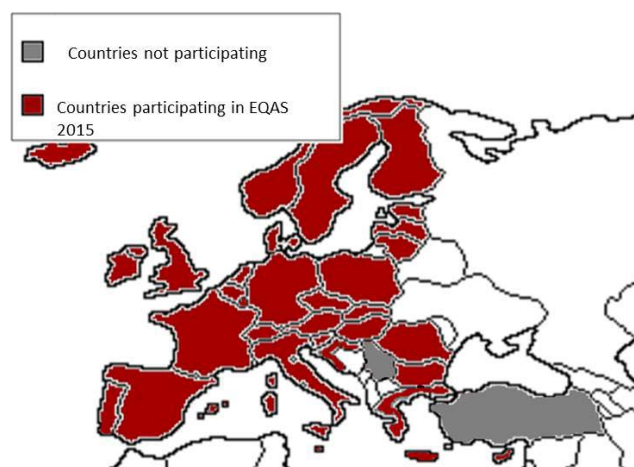


Figure 1 Participating countries in susceptibility testing of Enterococci, staphylococci and/or *E. coli*



Iceland (App. 2).

In total, this report includes AST results *E. coli* strains submitted by 31 laboratories, of enterococci strains submitted by 27 laboratories, of staphylococci strains submitted by 25 laboratories. The AST data included in the report represent all 28 MS in the EU (only one set of data per MS were included) and additionally includes data from laboratories in three non-EU countries (Norway, Switzerland and Iceland) (Figure 1).

2.2 Strains

Bacterial strains included in this EQAS (eight enterococci, eight staphylococci and eight *E. coli*) were selected among the DTU-Food strain collection on the basis of antimicrobial resistance profiles and previously obtained MIC values. For quality assurance purposes, one strain per each bacterial species tested has been included in all EQAS iterations performed to date, which represents an internal control.

AST of the EQAS strains was performed at DTU-Food by MIC determination using the Sensititre panels from Trek Diagnostic Systems. The MIC values obtained (App. 3) were used as reference values for this EQAS trial after verification performed by the U.S. FDA. Results from the following antimicrobials were however not verified by FDA: ampicillin and teicoplanin for enterococci; meropenem, colistin, temocillin and ertapenem for *E. coli* and sulfamethoxazol, tiamulin and trimethoprim for staphylococci. After comparison and verification of the MIC values obtained at DTU-Food and FDA, the strains were inoculated in agar as stab cultures, tested another time for AST and additionally for homogeneity at the DTU-FOOD laboratory, and dispatched to the participating laboratories on the 8th June 2015.

Reference strains *E. faecalis* ATCC 29212, *S. aureus* ATCC 29213 and *E. coli* ATCC 25922 were provided to new participating laboratories with instructions to store and maintain them for quality assurance purposes and future EQAS trials.

2.3 Antimicrobials

The panels of antimicrobials recommended for AST in this trial are listed in Table 1.

The antimicrobials tested were similar to the previous year and adjusted to the EU regulation EC652/2013 and in the case of staphylococci to the most recent EFSA recommendations.

Guidelines for performing AST were set according to the Clinical and Laboratory Standards Institute (CLSI) document – M7-A10 (2015) “Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard - tenth Edition” and whenever commercial methods were used, the guidelines of the manufacturer should be followed.

MIC results were interpreted by using EUCAST epidemiological cut-off values (www.eucast.org), as included in the regulation referred above or as recommended by EFSA and described in the EQAS protocol (App. 4). Results of the ESBL confirmatory testing were interpreted according to the recommendations by EFSA and as referred in the regulation, using MIC testing in the second panel of antimicrobials which is intended to be tested every time a strain was found resistant to either cefotaxime, ceftazidime or meropenem in the first *E. coli* panel and interpreted according to the protocol, enabling to conclude on the strain's presumptive ESBL/AmpC or carbapenemase phenotype.

Table 1. Panels of antimicrobials for susceptibility testing of bacteria included in this EQAS 2015 component

Enterococci	Staphylococci	<i>Escherichia coli</i> 1 st panel	<i>Escherichia coli</i> 2 nd panel
Ampicillin, AMP	Cefoxitin, FOX	Ampicillin, AMP	Cefepime, FEP
Chloramphenicol, CHL	Chloramphenicol, CHL	Azithromycin, AZI	Cefotaxime + clavulanic acid (F/C)
Ciprofloxacin, CIP	Ciprofloxacin, CIP	Cefotaxime, FOT	Cefotaxime, FOT
Daptomycin, DAP	Clindamycin, CLN	Ceftazidime, TAZ	Cefoxitin, FOX
Erythromycin, ERY	Erythromycin, ERY	Chloramphenicol, CHL	Ceftazidime, TAZ
Gentamicin, GEN	Gentamicin, GEN	Ciprofloxacin, CIP	Ceftazidime+ clavulanic acid (T/C)
Linezolid, LZD	Linezolid, LZD	Colistin, COL	Ertapenem, ETP
Quinupristin-dalfopristin (Synercid), SYN	Mupirocin, MUP	Gentamicin, GEN	Imipenem, IMI
Teicoplanin, TEI	Quinupristin-dalfopristin (Synercid), SYN	Meropenem, MERO	Meropenem, MERO
Tetracycline, TET	Sulfamethoxazole, SMX	Nalidixic acid, NAL	Temocillin, TRM
Tigecycline, TGC	Sulfamethoxazole+Trimethoprim, SXT	Sulfamethoxazole, SMX	
Vancomycin, VAN	Tetracycline, TET	Tetracycline, TET	
	Tiamulin, TIA	Tigecycline, TGC	
	Trimethoprim, TMP		
	Vancomycin, VAN		

2.4 Distribution

Protocols and all relevant information were uploaded on the EURL-AR website (<http://www.eurl-ar.eu>) thereby EQAS participants could access necessary information at any time. On the eighth of June 2015, the bacterial strains in agar stab cultures were dispatched in double pack containers (class UN 6.2) to the participating laboratories according to the International Air Transport Association (IATA) regulations as UN3373, biological substances category B.

2.5 Procedure

The participants were recommended to keep the agar stab cultures refrigerated until performance of AST according to the information posted on the EURL-AR website

(App. 4b, 4c, 4d and 4e). In addition, instructions for interpretation of AST results were provided. For interpretation of MIC determination results, cut-off values were reported in the protocol (App. 4b: Tables 1, 2 and 3). The EQAS test strains should be categorized as resistant or susceptible for every antimicrobial that has an available ECOFF.

The EURL-AR is aware that there are two different types of interpretative criteria of results, clinical breakpoints and epidemiological cut-off values. The terms 'susceptible', 'intermediate' and 'resistant' should be reserved for classifications made in relation to the therapeutic application of antimicrobial agents. When reporting data using epidemiological cut-off values, bacteria should be reported as 'wild-type' or 'non-wild-type' (Schwarz et al., 2010).

To simplify the interpretation of results, throughout this report, we will still maintain the terms susceptible and resistant, even in cases where we are referring to wild-type and non-wild-type strains.

All participating laboratories were invited to enter the obtained results into an electronic record sheet at the EURL-AR web-based database designed for this trial through a secured individual login and password.

A record sheet was provided with the protocol, including space for reporting the results (MIC values in mg/L) obtained for the reference strains. These results were compared to the quality control ranges reported by CLSI in

documents VET01 A4 (2013) / M100-S25 (2015) (App. 5).

The database was finally closed and evaluations were made available to participants on the 17th September 2015.

After this date, the participants were invited to login again to retrieve a database-generated individual report which contained an evaluation of the submitted results including possible deviations from the expected interpretations. Finally, participants were encouraged to complete an evaluation form available at the EURL-AR database with the aim to improve future EQAS trials

3. Results

The participants were asked to report results, including MIC values together with the categorisation as resistant or susceptible. Only the categorisation was evaluated, whereas the MIC values were used as supplementary information.

As mentioned in the introduction, the EURL-AR network established that data should be examined and possibly subtracted from the general analysis if there are less than 75% correct results for a strain/antimicrobial combination in the ring trial. In this respect, we have noticed in the raw data analysis at database closing that six antimicrobial/strain combinations were causing 25% or more deviations and these were further analysed in this report, and/or excluded from the analysis if this was justified. This was the case for strain ENT 9.3/tigecycline (29%), ENT 9.4/daptomycin (90%) and ENT 9.4/Quinopristin/dalfopristin (33%) as well as ST9.4/ciprofloxacin (58%), and ST9.5/Quinopristin-dalfopristin (SYN) (56%), EC 9.1/meropenem (33%).

After these results were analyzed, the results for the enterococci and staphylococci

combinations were deleted from the report as the cause for these deviations was that the expected values were just one step from the breakpoint. Thus, these tests results were not considered representative of the capacity of the laboratories for performing AST and were therefore not included in the report and the percentages of deviations for the participants were recalculated and presented in this report. However, in the case of the results of the combination EC 9.1/meropenem, the results were kept in the report given the importance of carbapenemase detection. This is due to the difficulties that arise in the of the phenotype of the OXA-48 gene, present in this strain. In the monitoring it is considered that the NRLs should be able to detect it even though it leads to reduced susceptibility to carbapenems which is at a rather low level and therefore it is challenging to detect this resistance.

3.1 Methods

As mentioned previously, all results were reported using MIC methods as described in the regulation. Furthermore, the database was designed only to receive data from MIC tests

Table 2. Total number of antimicrobial susceptibility tests (AST) performed for each EQAS 2015 strain and percentage (%) of correct results

Strain*	No. AST	No correct	% correct	Strain*	No. AST	No correct	% correct	Strain*	No. AST	No correct	% correct
ENT-9.1	278	273	98.2%	ST-9.1	316	314	99.4%	EC 9.1	591	557	94.2%
ENT-9.2	279	275	98.6%	ST-9.2	313	310	99.0%	EC 9.2	431	429	99.5%
ENT-9.3	255	254	99.6%	ST-9.3	314	312	99.4%	EC 9.3	641	637	99.4%
ENT-9.4	257	254	98.8%	ST-9.4	288	272	94.4%	EC 9.4	646	641	99.2%
ENT-9.5	296	290	98.0%	ST-9.5	295	291	98.6%	EC 9.5	646	641	99.2%
ENT-9.6	279	276	98.9%	ST-9.6	314	309	98.4%	EC 9.6	418	407	97.4%
ENT-9.7	277	272	98.2%	ST-9.7	315	313	99.4%	EC 9.7	649	649	100.0%
ENT-9.8	278	276	99.3%	ST-9.8	314	313	99.7%	EC 9.8	646	642	99.4%

*ENT, enterococci; ST, staphylococci; EC, *Escherichia coli*.

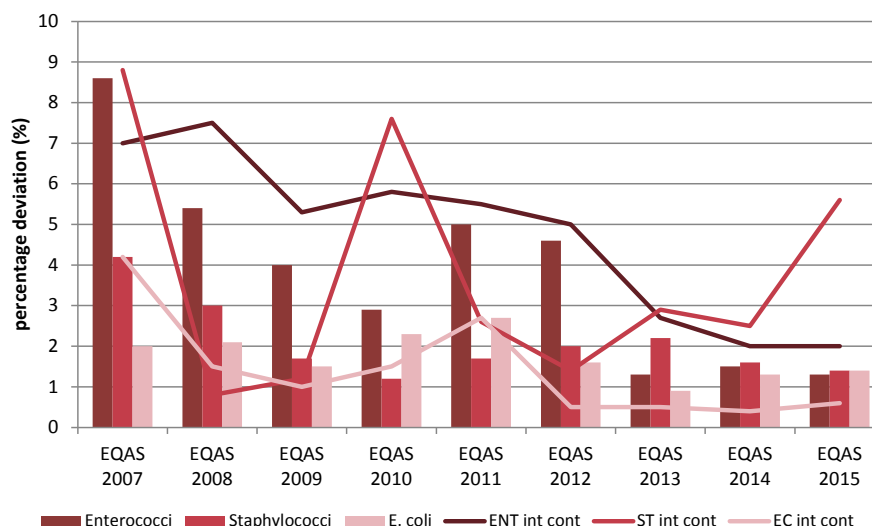


Figure 2 Overview of the percentages of deviations from expected results obtained in different EQAS iterations for the three bacterial species tested. The internal control strain is represented by a line.

including values and interpretations as well as QC data from MIC relevant strains.

In the EQAS 2015, 27, 25 and 31 participants performed AST by MIC determination for enterococci, staphylococci and *E. coli*, respectively.

3.2 Deviations overall

The list of deviations is illustrated in Appendices 8a, 8b and 8c. Figure 2 shows the overall

deviation levels.

Overall, the percentage of results in agreement with the expected values ranged from a minimum of 94.4% (strain ST 9.4) to a maximum of 100% (strain EC 9.7), as shown in Table 2. The *Enterococci* trial resulted in the highest percentage of correct results, which was at 98.7%, whereas it was only very slightly lower for both *E. coli* and staphylococci at 98.6% in general terms. The results for the

internal control strains were at similar levels as in 2014 for the *Enterococcus* (2.0%; same as 2014) and *E. coli* strains (0.6% vs 0.4% in 2014), but higher than previously for the *Staphylococcus* internal control strain (5.6% vs 2.5% in 2014) (Figure 2).

Detailed analyses of the results obtained for each species are reported in the following subchapters.

3.2.1 Enterococci

Analysis of results from the Enterococci trial showed that three antimicrobial/strain combinations had more than 25% deviations due to expected MIC being one dilution step from the breakpoint. This was the case of combination EURL ENT 9.3 and tigecycline, the combination ENT 9.4 and daptomycin and ENT 9.4 and quinopristin/dalfopristin (SYN). For ENT 9.3 and tigecycline, 21 laboratories uploaded results and of these 29% (n=6) have obtained a deviation by considering it as resistant. This strain had an expected result of “S” and an expected MIC value of 0.25 mg/L which is just below the breakpoint. All six laboratories having a deviation had submitted a MIC value of 0.5 mg/L (just one step above).

Similarly for ENT 9.4/daptomycin 22 laboratories uploaded results and from these a 20 (91%) considered the strain as susceptible while it was expected interpretation was set as resistant. The expected value of the MIC was 8 mg/L and 10 participants found the MIC to be at 4 mg/L while 2 observed it at 2 mg/L and only one at 1 mg/L.

The third combination of enterococci which was found problematic was ENT 9.4 and Quinopristin/dalfopristin with a deviation level at 33%. In this case, 21 results were uploaded and from these, seven were found deviating due to finding the MIC at 8 mg/L and interpreting it as “R” which was just one step deviation from the expected MIC at 4 mg/L and corresponding susceptible interpretation

whereas the remaining fourteen had the correct interpretation obtained with either observing and MIC of 4 mg/L in thirteen cases or 2 mg/L in one case.

These results were omitted from the calculations in this report as they do not reflect the capacity of the laboratories to perform AST.

Twenty-seven laboratories, representing 27 countries (24 MS and three non-EU countries) uploaded results for the enterococci trial. The Enterococci trial had in general excellent results with 98.7% of the AST results interpreted correctly.

Results deviating from expected interpretation subdivided by strain showed that the percentage of deviations ranged from 0.4% (ENT 9.3) to 2.0% (ENT 9.5) (Figure 3).

Analysis of the results according to the tested antimicrobials showed that the highest percentages of deviation from expected interpretations were obtained in testing susceptibility to tigecycline (11.0%) and quinopristin-dalfopristin (4.8%) (Figure 4). An overview of obtained and expected results is reported in Appendix 7a.

Enterococci identification (ID)

As a mandatory component of the proficiency test, the participants were requested to identify the enterococci species. The eight strains included six *E. faecalis* strains ENT 9.1, 9.2, 9.3, 9.6, 9.7 and 9.8) and two *E. faecium* (ENT 9.4 and 9.5). The results were excellent as no deviations were observed in the 208 results uploaded. However Lab # 38 did not upload results for the ID of the eight test strains as they referred in the comments that they did not do the species ID and therefore did not upload an interpretation of the results obtained for quinopristin/dalfopristin for any of the test strains.

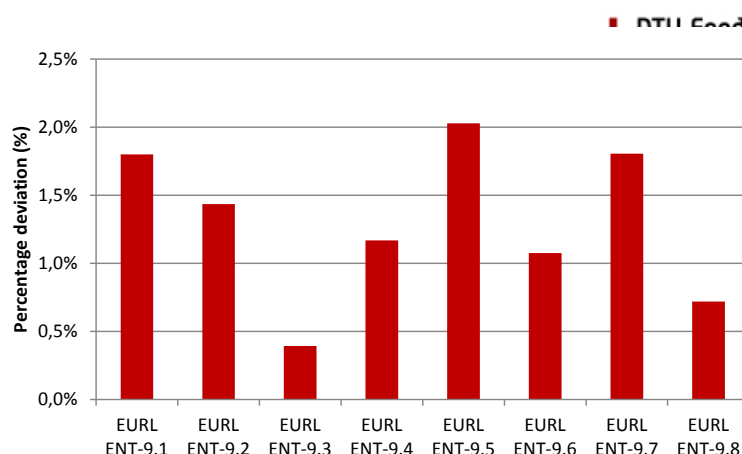


Figure 3. Enterococci trial: results deviating from the expected interpretation subdivided by tested strain

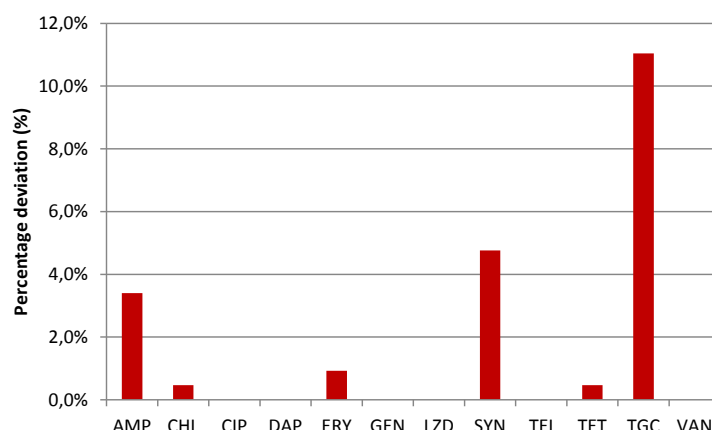


Figure 4. Enterococci trial: results deviating from the expected interpretation according to tested antimicrobials.

3.2.2 Staphylococci

The results of the staphylococci trial showed that there were more than 25% deviations for two antimicrobial/strain combinations due to expected results being very close to the breakpoint. These combinations were: ciprofloxacin/ST 9.4 (58%) and quinopristin-dalfopristin (SYN)/ ST 9.5 (56%).

For the combination ciprofloxacin/ST 9.4, 24 laboratories uploaded results and 58% of them (n=14) responded that the strain fell into the category of susceptible. This strain had an expected result of “R” due to an expected MIC value of 2 mg/L which is just above the breakpoint. From the fourteen laboratories having a deviation, ten laboratories observed an MIC value of 1mg/L (one step below the expected value), one laboratory had obtained a MIC value at 0.5 mg/L which is 2 steps below

the expected value and one additional laboratory obtained a MIC value at ≤ 0.25 mg/L.

Analysing the combination quinopristin dalfopristin/ST 9.5, we observed that a total of , 18 laboratories uploaded results and 56% of them (n=10) responded that the strain was resistant. This strain had an expected MIC at 1mg/L and was expected to be classified as susceptible. Nine of the laboratories reporting a deviation observed an MIC one step above the expected (at 2 mg/L), and one laboratory obtained an MIC two steps higher, at 4mg/L.

The results obtained for the three described antimicrobial/strain combinations were omitted from the calculations in this report as they did not reflect the capacity of the laboratories to perform AST.

Twenty-five laboratories representing the NRLs

for twenty-two MS and three non-MS countries have participated uploading results for the staphylococci trial.

In general the results showed more variation in the methods, breakpoints and antimicrobials tested as testing of *S. aureus* is not included in the EU regulation and therefore does not have fixed test panels, even though the technical specifications from EFSA are giving guidance to the tests to perform AST on this species. However, for the test results submitted, 98.6% had correct interpretations.

The results subdivided by strain showed that the obtained interpretations deviated from the expected ones slightly differently, with deviation percentages ranging from 0.3% for strain ST 9.8 to 5.6% for strain ST 9.4 (Figure 5). In this case, the strain ST 9.4 showed a larger percentage of deviations in relation to past trials. By this we mean that particular strain was the internal control strain and therefore we can compare to historical results which show that the deviation percentage for this strain was below 5% since 2011. The relatively high deviation percentage was due to sixteen deviations (after excluding the results omitted for ciprofloxacin, for this strain) from which fourteen are related to MIC measured too high by five different laboratories and affecting results of several antimicrobials.

Analysis of the staphylococci results sorted according to the tested antimicrobials showed that the highest percentages of deviation from expected interpretations were obtained in testing susceptibility to quinupristin-dalfopristin (SYN) (4.0%), followed by clindamycin (CLN) (3.7%) and erythromycin (ERY) (3%) (Figure 6).

An overview of obtained and expected results is reported in Appendix 7b.

Methicillin-resistant *S. aureus*

this EQAS trial, we had six strain expected to be identified as methicillin resistant: ST: 9.1,

9.2, 9.3, 9.4, 9.6 and 9.8. Strain ST 9.6 harboured the *mecC* gene and all the remaining harboured the *mecA* gene.

As for the EQAS on AST, all 25 participants on the staphylococci trial have submitted the methicillin results as these are a compulsory part the trial.

Only one laboratory (Lab #2) had three deviations in this trial due to reporting strains ST 9.1, 9.2 and 9.8 as methicillin susceptible. All remaining results were correct.

3.2.3 *Escherichia coli*

In the *E. coli* trial we also observed one strain/antimicrobial combination which showed more than 25% deviations. This was the case for meropenem EC 9.1. This strain EC 9.1 was exactly the same as the one sent out in 2014 as strain EC 8.7 and contains an OXA-48 gene conferring reduced susceptibility to carbapenems, without causing high levels of cephalosporin resistance. Therefore, among the 31 laboratories uploading results for this strain for the first panel, 14 (45.2%) of them considered this strain as susceptible in the first panel. However, not only the 17 participants that had found meropenem resistant but additional 7 participants (of the total of 24 participants) tested this strain in the second panel. In this test there were four participants (17.4%) that did not detect the meropenem resistance. This way, the meropenem testing for both panels had an average deviation level of 33.3%. The deviation level for imipenem tests in the second panel was quite low, (at 9%) unlike in the previous EQAS. These issues might not reveal real problems in the AST methodology, but issues related to the low level of resistance of the particular strain, as the strain's expected MIC for meropenem was relatively close to the breakpoint (0.5 mg/L) and most laboratories having this mistake had results just one step below the breakpoint at 0.12 mg/L, however four laboratories had MIC levels at 0.06 mg/L and one at 0.03 mg/L which

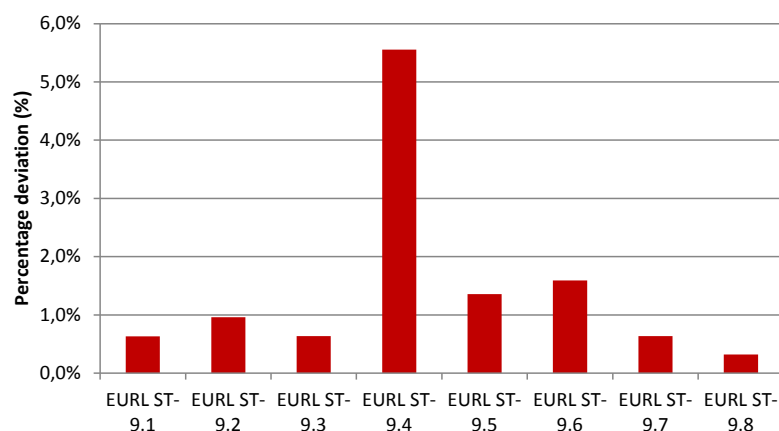


Figure 5. Staphylococci trial: results deviating from the expected interpretation subdivided by tested strain.

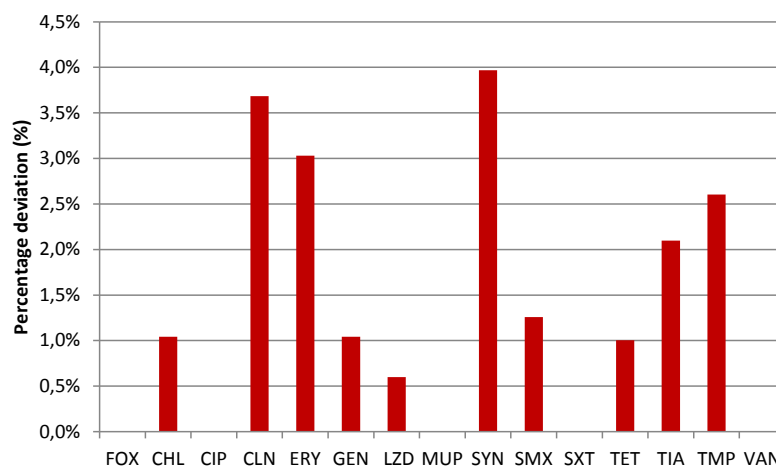


Figure 6. Staphylococci trial: results deviating from the expected interpretation according to tested antimicrobials.

could indicate loss of plasmid. There was also one laboratory where the interpretation was correct as “R”, even though the strain was tested at 0.12 mg/L for panel 1 as it probably was adjusted when finding a higher MIC value in panel 2, as well as several laboratories that performed panel 2 even though they did not observe meropenem resistance on the panel 1 results.

In general, the analysis of results from the *E. coli* trial showed that 98.6% of the results were interpreted correctly. Figure 2 shows the total percentage of deviations assigned to AST in this trial in relation to the previous trials which

is a very similar levels in relation to 2014.

Analysis of results deviating from expected interpretation subdivided by strain showed that the percentage of deviations ranged from none to 5.8% (Figure 7). The highest percentage of disagreement with expected results was obtained for EC 9.1 (Figure 7) and this is mainly due to the issues related to meropenem. An overview of obtained and expected results is reported in Appendix 7c.

The results sorted according to antimicrobials showed that the highest percentages of deviation from expected interpretations were obtained in testing susceptibility to cefepime



(5.1%) and meropenem (4.5%)(Figure 8). For cefepime the reason might be related mostly to cefepime expected MIC values close to the breakpoint in two of the strains EC 9.1 and 9.4 and for meropenem the explanations are given above as most deviations are related to strain EC 9.1 (Figure 8).

An overview of obtained and expected results is reported in Appendix 7c.

Beta-lactamase-producing *E. coli*

As for previous trials, the confirmation of beta-lactamase production is a mandatory component of this EQAS.

According to the protocol, which was based on the EFSA technical specifications the confirmatory test for ESBL production requires the testing of the second *E. coli* susceptibility testing panel. This panel is meant to be used to confirm the phenotype and perform a presumptive diagnosis of the type of gene(s) that might be present in the strains.

For identification of ESBL phenotypes, one of the main concepts is synergy which is defined as a ≥ 3 twofold concentration decrease in an MIC for either cephalosporin agent tested in combination with clavulanic acid vs. its MIC when tested alone (CLSI M100 Table 2A; Enterobacteriaceae). Resistance to cefepime gives further indication of ESBL production.

According to the most recent EFSA recommendations, confirmatory test for carbapenemase production requires the testing of meropenem (MER).

Detection of AmpC-type beta-lactamase producing bacteria can be performed by testing the isolates for susceptibility to ceftiofur (FOX). Resistance to FOX could indicate the presence of an AmpC-type beta-lactamase, that may be verified by PCR and sequencing.

The classification of the phenotypic results should be based on the most recent EFSA

recommendations (EFSA, 2012), indicating as:

- Presumptive ESBL: strains with positive synergy test, susceptible to ceftiofur and resistant to cefepime
- Presumptive ESBL+pAmpC: -strains with positive or negative synergy test, resistant to ceftiofur and resistant to cefepime
- Presumptive pAmpC phenotype: strains with negative synergy test resistant to ceftiofur and susceptible to cefepime
- Presumptive carbapenemase phenotype: strain resistant to meropenem
- Unusual phenotype: any other combinations

In this EQAS, 31 laboratories have uploaded results from which 30 uploaded results for all eight strains and one did not upload a result for strain EC 9.6.

Please note that for strain EC 9.4 no deviations were obtained as we adjusted the evaluation to accept both options "Presumptive pAmpC" and "presumptive ESBL+pAmpC" mixed phenotype obtained, even though this strain was an AmpC carrying CMY-2 and TEM-1B genes, but showing FEP resistance.

Deviations from expected results were obtained as follows:

Eight participants (Lab #17, #19 and #21, #23, #26, #29, #40 and #42) did not identify EC 9.1 as a carbapenemase producing strain as they did not find the carbapenemase resistance and classified it as: "No ESBL, AmpC- or carbapenemase.

One participant (Lab #56) misclassified EC 9.3 which was an ESBL carrying the CTX-M-1 gene

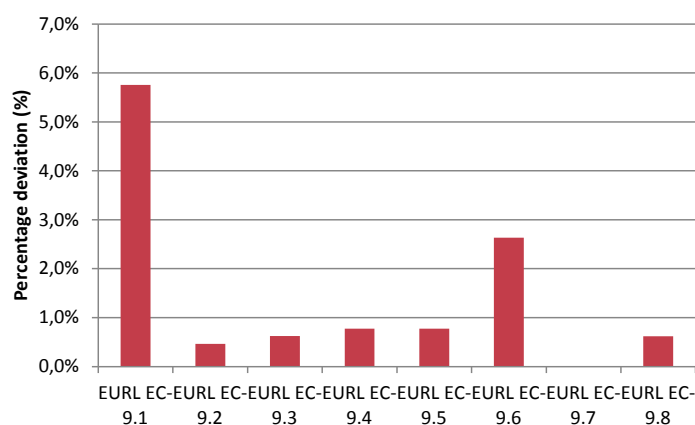


Figure 7 *E. coli* trial: results deviating from the expected interpretation subdivided by tested strain and antimicrobial susceptibility test method used

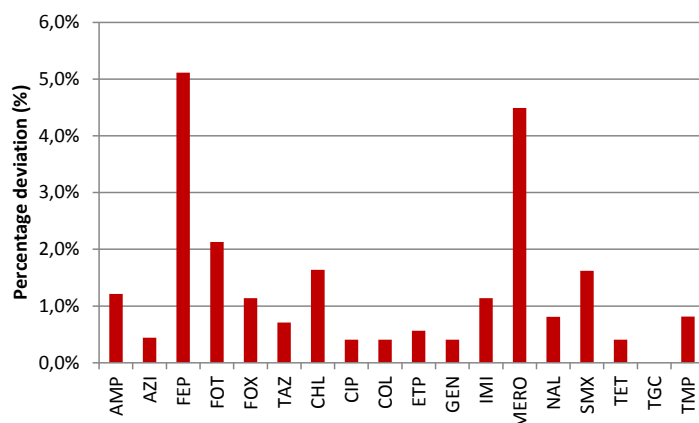


Figure 8. *E. coli* trial: results deviating from the expected interpretation according to tested antimicrobials.

as containing “No ESBL, AmpC- or carbapenemase”. Strain EC 9.6 did not harbour a resistant phenotype was misclassified as presumptive ESBL by Lab #33 and presumptive pAmpC by lab #39. This was not further explained but could be related to switch of trains or results as further results were found deviating for this strain.

Regarding the EC 9.7 this strain had a mixed phenotype due to the CTX-M-1 and CMY-2 genes and was expected to be classified as presumptive ESBL+pAmpC. Only one deviation was noted as Lab #30 considered this strain only to be presumptive ESBL.

Finally, strain EC 9.8 was an ESBL strain harbouring TEM-52 and only one participant (Lab#19) had a deviation due to classifying it as presumptive ESBL+AmpC.

3.3 Deviations by participating laboratory

The figures 9, 10 and 11 illustrate the percentage of deviations for each participant laboratory for each of the trials.

One out of 27 participants obtained a percentage of deviations from expected results higher than 5% for enterococci (Figure 9), one out of 25 participants had above 5% deviation in the staphylococci trial (Figure 10) and none out of the 31 participants had above 5%



deviation in the *E. coli* trial (Figure 11). These results will be the focus of the next sections.

3.3.1 Enterococci

The largest number of deviations (9.2%) and was obtained by lab #42. This laboratory reported all eight enterococci strains as resistant to tigecycline as they found the MIC at either 0.5 or 1 mg/L which is either one or two steps above the breakpoint. The expected MIC for the strains was 0.12 and 0.25 mg/L and were all expected to be reported as susceptible. As the deviation for the strain 9.3 was omitted, due to the high percentage level for this strain for the remaining participants, the tigecycline issue caused seven deviations for this participant after omission. Additionally, this participant had one deviation on strain ENT 9.5 for ampicillin. Follow-up on the issues showed that the tigecycline issue might be related to the sensitivity of this particular drug to oxygen and light which might have been related to the batch of plates and media used. As a follow up on this issue we have run some test at the EURL with the batches of EUVENC plates we managed to receive from the NRLs. We have received 28 plates from four laboratories (including Lab#42) and representing two batches: B4255 (18 plates) and B 4423A (10 plates). In the laboratory we performed the testing of all Enterococci from EQAS 2015 and the *E. faecalis* ATCC 29212 on all the combinations that were possible. Testing was performed using the same methodology in parallel and using the same Mueller Hinton broth from TREK (batch 656272).

As a result we observed that for all the tigecycline tests the results would be falling between 0.06 and 0.25 and interpreted as susceptible. As the deviations observed were due to obtaining MIC values at 0.5 or 1mg/L and interpreting the strain as resistant, we could not see this in our tests, so we cannot confirm that there was an issue with the plates

and we can only conclude that the observed deviations in the EQAS might be due to issues related to the methodology or the media used in the laboratories affected. Later on, in the troubleshooting performed by the participant of the Lab#42 a more concrete cause for the deviation was found as the panels were incubated for 48h, which does not comply with the standards. However, as reported to the EURL-AR this modification in the procedure had not been applied to any of the routine isoaltes, but only when processing the EQAS strains.

We do, therefore recommend to look closely at this issues and follow up on enterococci showing unexpected resistance to this antimicrobial which should be considered as a rare event.

The second laboratory having a percentage of deviation above 5% for was lab #12 at 7.1% which obtained four deviations for (ENT 9.1, 9.2, 9.3 and 9.5) for ampicillin due to higher MIC than the expected. In the follow up the participant mentioned that VETMIC plates were used and they were close to expiration date, which could explain the issues observed. Furthermore, this laboratory did not provide data for all the twelve antimicrobials, as they were using VETMIC plates and testing only to seven of these for each strain, thus influencing the number of test results. It was also clarified that this laboratory is not providing data for enterococci monitoring to EFSA but only running the EQAS strains and some other from diagnostic sources.

The third laboratory having more than 5% deviation was laboratory #19 that had five deviations leading to 5.7% and similarly to lab #42 the deviations were due to tigecycline tested with one step above the breakpoint and the expected MIC.

For further information please consult the

overview in the Appendixes (App. 8a).

In summary, 26 of the 27 participants in the enterococci trial achieved the acceptance level by having less than 5% of results deviating from the expected values (Figure 9).

3.3.2 Staphylococci

Analysis of laboratory performance of AST showed that one out of 25 participants obtained a percentage of deviations from expected results higher than 5.0% (Figure 10).

Participant #29 was considered outlier due to the percentages of deviations obtained. This participant had 9.1% deviations corresponding to ten deviations (there were initially 12 deviations but as two initial deviations were relating to strain/antimicrobial combinations omitted from the report these are not mentioned further). These deviations were related to the testing of three strains; ST 9.4 (six antimicrobials), ST 9.5 (two antimicrobials) and ST 9.6 (two antimicrobials). For most of the deviations higher MICs were obtained for the test strains than expected. After communication with the participant it was referred that the deviations were caused by Mueller Hinton broth that was not prepared according to the requirements. Furthermore, the participant reported that measures had been implemented to ensure performance of employees preparing media.

In summary, 24 of 25 participants in the staphylococci trial achieved the acceptance level by having less than 5% of results deviating from the expected values and only one laboratory had a larger deviation percentage and was considered an outlier (Figure 10).

Deviations from expected results obtained by each participant in the staphylococci trial are reported in Appendix 8b.

3.3.3 Escherichia coli

Analysis of laboratory performance of AST showed that none of the 31 participants obtained a percentage of deviations from expected results higher than 5% in the *E. coli* trial, being the highest deviation level at 4.5% and in this way all participants in the *E. coli* trial achieved the acceptance level by having less than 5% of results deviating from the expected values (Figure 12).

3.4 Deviations from expected results for the reference strains

The results for antimicrobial susceptibility testing of the reference strains have been evaluated according to the CLSI-established quality control (QC) ranges (App. 5).

3.4.1 Enterococcus faecalis ATCC 29212

In total 27 participants performed AST of *E. faecalis* ATCC 29212 by MIC determination. Three of the results for tigecycline were found outside of range due to obtained MICs one or two steps too high. This was the case for the results submitted by participant labs #2, #19 and #42 which also had deviations in the results uploaded for the test strains showing the same trend.

In summary, out of 299 tests performed by 27 participants, 296 test results were found correct (Table 3).

3.4.2 Staphylococcus aureus ATCC 29213

Twenty-five participants performed AST of *S. aureus* ATCC 29213 by MIC determination towards a variable number of antimicrobials. In this EQAS only one deviation was obtained by Lab #37 by testing sulfamethoxazole one step below the QC range (Table 4). In summary, out of 276 tests submitted, 275 were correct and one was deviating.

3.4.3 Escherichia coli ATCC 25922

Thirty-one participants performed AST of *E. coli* ATCC 25922 by MIC determination and

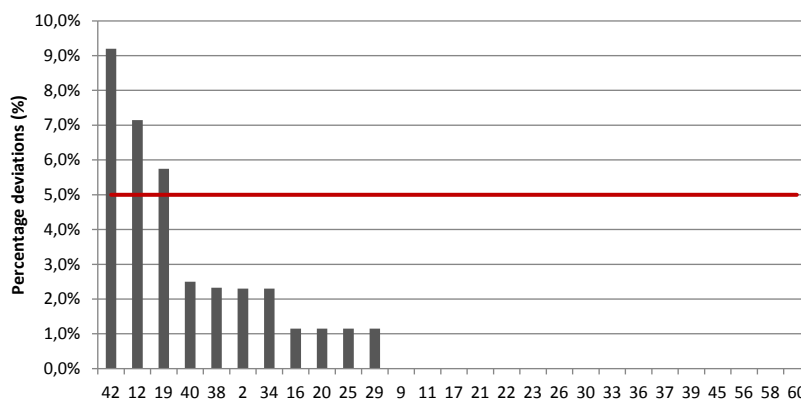


Figure 9 Percentage of deviations from expected results obtained by each laboratory in the Enterococci trial. The laboratories were ranked by decreasing percentage of deviations from expected results in antimicrobial susceptibility testing.

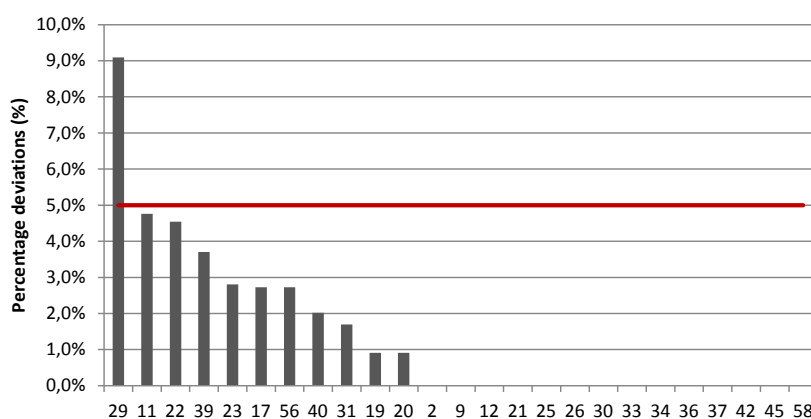


Figure 10 Percentage of deviations from expected results obtained by each laboratory in the Staphylococci trial. The laboratories were ranked by decreasing percentage of deviations from expected results in antimicrobial susceptibility testing.

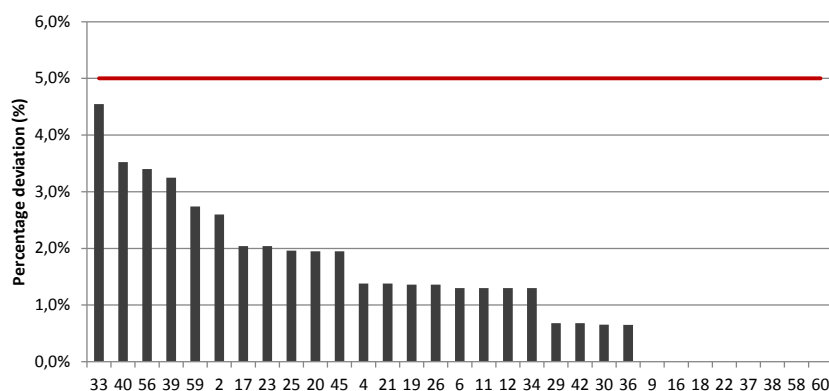


Figure 11. Percentage of deviations from expected results obtained by each laboratory in the *Escherichia coli* trial. The laboratories were ranked by decreasing percentage of deviations from expected results in antimicrobial susceptibility testing

uploaded data for the reference strain regarding the antimicrobials in the first panel for which there is a QC range available whereas only 27 uploaded values for the second panel. One value for sulfamethoxazole was not reported correctly by lab #59 as “###” was reported

Table 3. Antimicrobial susceptibility testing of *Enterococcus faecalis* ATCC 29212 by MIC determination: deviations from expected values.

Antimicrobial	Proportion outside of range	Below QC range	Above QC range
Ampicillin	0/26 (0%)	-	-
Chloramphenicol	0/27 (0%)	-	-
Ciprofloxacin	0/24 (0%)	-	-
Daptomycin	0/22 (0%)	-	-
Erythromycin	0/27 (0%)	-	-
Gentamicin	0/27 (0%)	-	-
Linezolid	0/27 (0%)	-	-
Quinu-dalfo-pristin	0/21 (0%)	-	-
Teicoplanin	0/22 (0%)	-	-
Tetracycline	0/27 (0%)	-	-
Tigecycline	3/22 (14%)	-	1 step (2) 2 steps (1)
Vancomycin	0/27 (0%)	-	-

Table 4. Antimicrobial susceptibility testing of *Staphylococcus aureus* ATCC 29213 by MIC determination: deviations from expected values.

Antimicrobial	Proportion outside of range	Below QC range	Above QC range
Cefoxitin	0/23 (0%)	-	-
Chloramphenicol	0/24 (0%)	-	-
Ciprofloxacin	0/24 (0%)	-	-
Clindamycin	0/24 (0%)	-	-
Erythromycin	0/25 (0%)	-	-
Gentamicin	0/24 (0%)	-	-
Linezolid	0/20 (0%)	-	-
Mupirocin	No range	-	-
Quinu-dalfo-pristin	0/19 (0%)	-	-
Sulfisoxazole	1/20 (5%)	1 step	-
Sulfamethoxazol + Trimethoprim	0/4 (0%)	-	-
Tetracycline	0/25 (0%)	-	-
Tiamulin	No range	-	-
Trimethoprim	0/24 (0%)	-	-
Vancomycin	0/20 (0%)	-	-

instead of a valid MIC value. Four deviations were detected by the automatic system, however one was due to entering “###” instead of a valid MIC value and therefore cannot count as a deviation on the testing. The remaining three deviations were obtained for nalidixic acid tested one step above range (Lab#19), sulfamethoxazole tested one step above range (Lab #16) and trimethoprim tested one step below the QC range (Lab #60). For the antimicrobials tested in the second panel there were some laboratories that did not upload data for testing the reference strain on this panel. This was the case for Labs #22, #26, 37 and #60. Additionally and #21 uploaded partial data lacking cefotaxime and ceftazidime results however all results uploaded were found within range.

In summary, out of 402 tests performed in the first panel, 398 were correct and in the second panel all 187 tests performed were found correct.

For further information please consult App 6a, 6b and 6c.

Table 5. Antimicrobial susceptibility testing of *Escherichia coli* ATCC 25922 by MIC: deviations from expected values.

Antimicrobial	Panel	Proportion outside of range	Below QC range	Above QC range
Ampicillin	1	0/31 (0%)	-	-
Azithromycin	1	No range	-	-
Cefotaxime	1	0/30 (0%)	-	-
Ceftazidime	1	0/31 (0%)	-	-
Chloramphenicol	1	0/31 (0%)	-	-
Ciprofloxacin	1	0/31 (0%)	-	-
Colistin	1	0/31 (0%)	-	-
Gentamicin	1	0/31 (0%)	-	-
Meropenem	1	0/31 (0%)	-	-
Nalidixic acid	1	1/31 (3%)	-	1 step
Sulfamethoxazole	1	2/31 (6%)	####*	1 step
Tetracycline	1	0/31 (0%)	-	-
Tigecycline	1	0/31 (0%)	-	-
Trimethoprim	1	1/31 (3%)	1 step	-
Cefepime	2	0/27 (0%)	-	-
Cefotaxime	2	0/26 (0%)	-	-
CTX/clav acid	2	No range	-	-
Cefoxitin	2	0/27 (0%)	-	-
Ceftazidime	2	0/26 (0%)	-	-
CAZ/ clav acid	2	No range	-	-
Ertapenem	2	0/27 (0%)	-	-
Imipenem	2	0/27 (0%)	-	-
Meropenem	2	0/27 (0%)	-	-
Temocillin	2	No range	-	-

- One participant wrote "####" into the response field, therefore obtaining a deviation, which cannot be considered a real mistake.

4. Discussion

4.1 General overview

In general, the results were comparable to recent years and the overall deviation levels for AST in the three trials were just slightly lower for enterococci and staphylococci and slightly higher for *E. coli* with percentage of deviations ranging from 1.3% to 1.4%. (Figure 2). The results observed with the internal control strain, showed similar value for the enterococcus strain and higher deviation level for the staphylococcus strain and a smaller increase in the deviation level for the *E. coli* strain. The results for all three bacterial species and the deviation levels for these strains ranged from 0.6% to 5.6% (Figure 2).

It is important to consider that the number of EQAS participants changes slightly from year to year, which implies that comparisons among different EQAS iterations might be difficult to interpret. Furthermore, results from three laboratories from EU-affiliated countries non-MS were included in this report.

The network has now implemented the EU regulation and therefore the AST methodology has been harmonized among NRLs for testing *E. coli* and enterococci. This shows by having most laboratories uploading data for all antimicrobials in the panels. However, not all results are uploaded, denoting possibly that not all laboratories are yet able to deliver data for all antimicrobials. However, as staphylococci

are not included in the regulation there are some discrepancies in the tests performed in relation to the EFSA recommended antimicrobials, between participants as they are using different panels for the testing.

4.2 Enterococci

The percentages of deviations observed ranged from 0.4% to 2.0% among the different test strains (Figure 3). These percentages are relatively similar to previous trials in the latest years.

As mentioned previously, three participants submitted more than 5% deviating results (Figure 9). These participants have been contacted by the EURL-AR to perform troubleshooting to find the possible causes of issues in the performance and as problems have been detected regarding tigecycline the EURL also obtained panel from the possibly affected batches to compare. Results have been disseminated to the NRL network and further discussions on this subject will be included in the forthcoming EURL-AR Workshop. When comparing the 2015 results with 2014, the deviation level was actually decreased in total.

The number of participants performing AST for enterococci with 100% agreement with the expected results was 16 (59%), which is only slightly fewer than last year.

The results for the of the quality control strain *E. faecalis* ATCC 29212 was very good for the 27 participants (Table 3). In summary, out of 299 tests performed overall, 296 (98.9%) were within range and only three were deviating.

Regarding the identification of the enterococci strains, the results were excellent with no deviations in the 208 tests performed. However one laboratory (Lab #38) did not upload species identification results.

4.3 Staphylococci

The deviation percentages observed among the

results for the different test strains ranged from 0.3% to 5.6% among the different test strains which is slightly higher than in previous trials (Figure 5). However, the number of participants performing AST with 100% agreement with the expected results was higher than in the past trials and consisted of 14 participants (56%).

Identification of methicillin-resistant strains was in general very good as only one laboratory (Lab #2) obtained all the three deviations observed in this trial, meaning that the remaining 24 laboratories had 100% correct methicillin resistance detection, which includes both *mecA* and *mecC*. This also means that laboratories within the EURL-AR network in general correctly identify MRSA. The deviations obtained by Lab #2 are related to two false negatives and one false positive result. From a total of 200 submitted results 197 (98.5%) were correct.

AST of the quality control strain *S. aureus* ATCC 29213 in MIC determination resulted in 99.6% correct tests as from the 276 submitted test results only one was deviating (Table 4). Overall, this performance was quite satisfactory.

4.4 Escherichia coli

The percentages of results deviating from the expected interpretations varied from 0.3% to 6.9% among the different test strains, with seven of the strains showing deviation percentages between 0% to 5.8% and mostly strain EC 8.7 had a high deviation percent due to the difficulties observed in detection of meropenem resistance in both test panels (Figure 7). For further detail in the deviations observed please consult Appendix 8c.

None of the 31 participants had deviation percentages above the 5% acceptance level, which is remarkable (Figure 11).

The number of participants performing AST with 100% agreement with the expected results was

however not very high at 8 (25%). Again, as for the previous year, this is mainly due to the high number of deviations for meropenem results for strain EC 9.1.

As the results show for the third time, the detection of OXA- type carbapenemases should be further improved as eight deviations were noticed in classifying the strain EC 9.1.

Additionally, we consider there is still room for improvements in the performance and interpretation of ESBL and AmpC phenotypes

as five deviations were found concerning both ESBL and AmpC phenotype classification.

AST of the quality control strain *E. coli* ATCC 25922 resulted in 99% correct tests for both the first panel and the second panel as from a total of 589 tests, 585 were correct and four deviating, from which only three were real deviations (Table 5). Overall, this performance was quite satisfactory.

5. Conclusions

In 2015, the number of laboratories not performing AST above the acceptance level (i.e. > 5% deviating results) was relatively low and consistent with the results obtained in previous EQAS trials. Three out of 27 participants obtained a percentage of deviations from expected results above 5% for enterococci (Figure 9), one out of 25 participants had above 5% deviation in the staphylococci trial and was considered an outlier (Figure 10) and none of the 31 participants were above the acceptance threshold in the *E. coli* trial (Figure 11).

The laboratories outside the acceptable level have been contacted to assess individually the causes of inadequate AST performance and these individual contacts should be taken as an opportunity to perform troubleshooting and self-evaluation and to discuss with the EURL-AR on how to improve the AST results in the future.

The enterococci ID module did not reveal any methodological issues, but as one participant did not upload this parameter, the EURL-AR will follow up on the laboratory capacity of performing the ID.

In this trial, few issues with the methicillin resistance results were reported and therefore EURL-AR will follow up on any needs regarding the implementation of the correct detection and

confirmation methods in these laboratories.

Major focus will again be given next year on the correct identification of *E. coli* producing beta-lactamases of the ESBL, AmpC and especially the OXA-48 phenotypes which are difficult to detect. Also as part of the testing of isolates coming from the selective isolation where a large number of suspect isolates need to undergo phenotypic screening and be selected for confirmatory testing these methods need to be applied correctly. We strongly encourage participants having difficulties in identifying these phenotypes to perform a re-test of the test strains as a training exercise, and to contact the EURL-AR in case any discussion is needed.

Furthermore, focus will be at the new important resistance mechanism discovered- plasmid mediated colistin resistance.

Finally, the EURL-AR welcomes suggestions for improvement in future EQAS trials and invites the network to contribute with ideas for material to be disseminated in newsletters, alert for training needs on specific focus areas which may be of interest of the network and improve the knowledge and skills of the laboratories involved in the AMR monitoring.

6. References

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November 2013 on the monitoring and reporting of antimicrobial resistance in zoonotic and commensal bacteria

Schwarz S, Silley P, Simjee S, Woodford N, van DE, Johnson AP & Gaastra W. (2010) Editorial: assessing the antimicrobial susceptibility of bacteria obtained from animals. J Antimicrob Chemother 65: 601-604.

Appendix 1- EURL-AR EQAS pre-notification

EQAS 2015 FOR *E. COLI*, STAPHYLOCOCCI AND ENTEROCOCCI

The EURL-AR announces the launch of another EQAS, thus providing the opportunity for proficiency testing which is considered an essential tool for the generation of reliable laboratory results of consistently good quality.

This EQAS consists of antimicrobial susceptibility testing of eight *E. coli* isolates, eight staphylococci and eight enterococci isolates. Additionally, quality control (QC) strains *E. coli* ATCC 25922 (CCM 3954), *E. faecalis* ATCC 29212 (CCM 4224) and *S. aureus* ATCC 29213 (CCM 4223) (for MIC) will be distributed to new participants.

This EQAS is specifically for NRL's on antimicrobial resistance. Laboratories designated to be NRL-AR do not need to sign-up to participate but are automatically regarded as participants. You may contact the EQAS-Coordinator if you wish to inform of changes in relation to your level of participation in previous years. The EURL-AR will be able to cover the expenses for one parcel, only, per EU Member State. Therefore, countries with more than one laboratory registered on the EURL-AR contact-list will be contacted directly to confirm which laboratory will be included for participation free of charge.

The invitation to participate in the proficiency test is extended to additional participants from official NRLs and participants from laboratories which are involved in the network but are not designated NRLs (cost for participation will be 100 euro).

TO AVOID DELAY IN SHIPPING THE ISOLATES TO YOUR LABORATORY

The content of the parcel is "UN3373, Biological Substance Category B. Eight *E. coli*, eight staphylococci, eight enterococci and for new participants also the QC strains mentioned above. Please provide the EQAS coordinator with documents or other information that can simplify customs procedures (e.g. specific text that should be written on the proforma invoice). To avoid delays, we kindly ask you to send this information already at this stage.

TIMELINE FOR RESULTS TO BE RETURNED TO THE NATIONAL FOOD INSTITUTE

Shipment of isolates and protocol: The isolates will be shipped in *June* 2015. The protocol for this proficiency test will be available for download from the website (www.eurl-ar.eu).

Submission of results: Results must be submitted to the National Food Institute **no later than September, 4th, 2015**, via the password-protected website.

Upon reaching the deadline, each participating laboratory is kindly asked to enter the password-protected website once again to download an automatically generated evaluation report.

EQAS report: A report summarising and comparing results from all participants will be issued. In the report, laboratories will be presented coded, which ensures full anonymity. The EURL-AR and the EU Commission, only, will have access to un-coded results. The report will be publicly available.

Next EQAS: The next EURL-AR EQAS that we will have is on antimicrobial susceptibility testing of *Salmonella* and *Campylobacter* and a new EQAS on isolation of ESBL and ampC –producing *E. coli* from samples which are both expected to be carried out in *October, 2015*.

Please contact me if you have comments or questions regarding the EQAS.

Sincerely,
Lina Cavaco,
EURL-AR

Appendix 2- List of participants

Institute	Country	E coli	Ent	Staph
Austrian Agency for Health and Food Safety	Austria	x	x	x
Institute of Public Health	Belgium	x		
Nacional Diagnostic and Research Veterinary Institute	Bulgaria	x	x	x
Croatian Veterinary Institut	Croatia	x	x	x
Veterinary Services	Cyprus	x		
State Veterinary Institute Praha	Czech Republic	x	x	x
Danish Veterinary and Food Administration	Denmark	x	x	
Estonian Veterinary and Food Laboratory	Estonia	x	x	x
Finnish Food Safety Authority EVIRA	Finland	x	x	x
Agence nationale de sécurité sanitaire ANSES - Fougères	France	x	x	
Federal Institute for Risk Assessment	Germany	x	x	x
Veterinary Laboratory of Chalkis	Greece	x		
Central Agricultural Office Veterinary Diagnostic Directorate	Hungary	x	x	x
University of Iceland	Iceland	x	x	x
Central Veterinary Research Laboratory	Ireland	x	x	x
Istituto Zooprofilattico Sperimentale delle Regioni Lazio e Toscana	Italy	x	x	x
Institute of Food Safety, Animal Health and Environment "BIOR"	Latvia	x	x	x
National Food and Veterinary Risk Assessment Institute	Lithuania	x	x	x
Laboratoire national de Santé	Luxembourg	x		
Public Health Laboratory	Malta	x	x	x
Food and Consumer Product Safety Authority (VWA)	Netherlands	x	x	x
Central Veterinary Institute of Wageningen UR	Netherlands	x	x	x
Veterinærinstituttet	Norway	x	x	x
National Veterinary Research Institute	Poland	x	x	x
Laboratorio Nacional de Investigação Veterinária	Portugal	x	x	x
Institute for Diagnosis and Animal Health	Romania	x	x	x
State Veterinary and Food Institute (SVFI)	Slovakia	x	x	x
National Veterinary Institute	Slovenia	x	x	x
Laboratorio Central de Sanidad, Animal de Santa Fe	Spain			x
Laboratorio Central de Sanidad, Animal de Algete	Spain	x	x	
VISAVET Health Surveillance Center, Complutense University	Spain	x	x	x
Agencia Espanola de Seguridad Alimentaria y Nutricion	Spain	x	x	
National Veterinary Institute, SVA	Sweden	x	x	x
Vetsuisse faculty Bern, Institute of veterinary bacteriology	Switzerland	x	x	x
The Veterinary Laboratory Agency	UK	x	x	x

NRL's	
non- NRL enrolled for EQAS or extra NRL enrolled	
not EU-member state	

Appendix 3a- Expected results for the enterococci trial (MIC- values and interpretations)

Strain nr	Species	DAP	TGC	TEI	AMP	CHL	CIP	ERY	GEN	LZD	SYN	TET	VAN
EURL ENT 9.1	<i>E. faecalis</i>	2	0,25	<=0,5	1	8	1	>128	>1024	2	32	64	2
EURL ENT 9.2	<i>E. faecalis</i>	1	0,25	<=0,5	1	128	32	>128	<=8	2	8	128	1
EURL ENT 9.3	<i>E. faecalis</i>	1	0,25	<=0,5	1	128	1	>128	256	2	16	128	1
EURL ENT 9.4	<i>E. faecium</i>	8	0,25	<=0,5	>64	8	4	>128	8	2	4	128	2
EURL ENT 9.5	<i>E. faecium</i>	0,5	0,12	>128	4	8	0,5	2	<=8	2	4	64	>128
EURL ENT 9.6	<i>E. faecalis</i>	2	0,25	<=0,5	1	8	1	>128	16	2	16	<=1	2
EURL ENT 9.7	<i>E. faecalis</i>	1	0,25	<=0,5	1	128	1	>128	8	2	16	128	2
EURL ENT 9.8	<i>E. faecalis</i>	4	0,25	<=0,5	1	8	1	<=1	16	2	8	<=1	4

Strain nr	Species	DAP	TGC	TEI	AMP	CHL	CIP	ERY	GEN	LZD	SYN	TET	VAN
EURL ENT 9.1	<i>E. faecalis</i>	S	S	S	S	S	S	R	R	S	NA	R	S
EURL ENT 9.2	<i>E. faecalis</i>	S	S	S	S	R	R	R	S	S	NA	R	S
EURL ENT 9.3	<i>E. faecalis</i>	S	S	S	S	R	S	R	R	S	NA	R	S
EURL ENT 9.4	<i>E. faecium</i>	R	S	S	R	S	S	R	S	S	S	R	S
EURL ENT 9.5	<i>E. faecium</i>	S	S	R	S	S	S	S	S	S	S	R	R
EURL ENT 9.6	<i>E. faecalis</i>	S	S	S	S	S	S	R	S	S	NA	S	S
EURL ENT 9.7	<i>E. faecalis</i>	S	S	S	S	R	S	R	S	S	NA	R	S
EURL ENT 9.8	<i>E. faecalis</i>	S	S	S	S	S	S	S	S	S	NA	S	S

	Resistant
NA	Not applicable

Abbreviations: DAP- daptomycin, TIG- tigecycline, TEI- teicoplanin, AMP-ampicillin, CHL-chloramphenicol, CIP- ciprofloxacin, ERY- erythromycin, GEN- gentamicin, LZD- linezolid, SYN- quinupristin-dalfopristin, TET- tetracycline, VAN- vancomycin

Appendix 3b- Expected results for the staphylococci trial (MIC- values and interpretations)

Strain nr	Species	VAN	SYN	LZD	MUP	CLN	CHL	CIP	ERY	FOX	GEN	SMX	SXT	TET	TIA	TMP	methicillin R
EURL ST 9.1	<i>S. aureus</i>	<=1	8	2	<=0.06	>256	8	0.25	>16	8	0.25	<=32	<=0.25	>32	>32	<=0.5	MRSA
EURL ST 9.2	<i>S. aureus</i>	<=1	2	2	<=0.06	4	8	0.12	0.25	8	0.25	<=32	<=0.25	>32	>32	>32	MRSA
EURL ST 9.3	<i>S. aureus</i>	<=1	2	2	<=0.06	8	8	8	0.25	8	0.5	<=32	0.5	>32	>32	>32	MRSA
EURL ST 9.4	<i>S. aureus</i>	<=1	<=0,5	2	<=0,06	0.12	8	2	0.25	8	>16	256	<=0.25	32	0.5	1	MRSA
EURL ST 9.5	<i>S. aureus</i>	<=1	1	2	<=0.06	0,5	8	0,25	0.5	4	0.5	<=32	<=0.25	0.5	>32	1	MSSA
EURL ST 9.6	<i>S. aureus</i>	<=1	<=0,5	4	<=0.06	0.25	8	0.5	0.5	8	0,25	<=4	<=0.25	0,5	1	1	MRSA
EURL ST 9.7	<i>S. aureus</i>	<=1	<=0.5	2	<=0,06	0.12	8	0.25	0.25	4	0.25	<=32	<=0.25	0.25	1	1	MSSA
EURL ST 9.8	<i>S. aureus</i>	<=1	8	2	<=0.06	>256	16	0.25	>16	8	0.5	<=32	0.5	>32	>32	>32	MRSA

Strain nr	Species	VAN	SYN	LZD	MUP	CLN	CHL	CIP	ERY	FOX	GEN	SMX	SXT	TET	TIA	TMP	methicillin R
EURL ST 9.1	<i>S. aureus</i>	S	R	S	S	R	S	S	R	R	S	S	S	R	R	S	MRSA
EURL ST 9.2	<i>S. aureus</i>	S	R	S	S	R	S	S	S	R	S	S	S	R	R	R	MRSA
EURL ST 9.3	<i>S. aureus</i>	S	R	S	S	R	S	R	S	R	S	S	S	R	R	R	MRSA
EURL ST 9.4	<i>S. aureus</i>	S	S	S	S	S	S	R	S	R	R	R	S	R	S	S	MRSA
EURL ST 9.5	<i>S. aureus</i>	S	S	S	S	R	S	S	S	S	S	S	S	S	R	S	MSSA
EURL ST 9.6	<i>S. aureus</i>	S	S	S	S	S	S	S	S	R	S	S	S	S	S	S	MRSA
EURL ST 9.7	<i>S. aureus</i>	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	MSSA
EURL ST 9.8	<i>S. aureus</i>	S	R	S	S	R	S	S	R	R	S	S	S	R	R	R	MRSA

	Resistant
NA	Not applicable

Abbreviations:, CHL- chloramphenicol, CIP- ciprofloxacin, CLN- Clindamycin, ERY- erythromycin, FOX- ceftiofur, LZD- linezolid, MUP- mupirocin, GEN- gentamicin, SYN- quinupristin-dalfopristin,, SMX- sulphamethoxazole, SXT- sulphamethoxazole + trimethoprim, TET- tetracycline, TIA- tiamulin, TMP- trimethoprim, VAN- vancomycin

Appendix 3c- Expected results for the E. coli trial (MIC- values and interpretations)

Panel 1

Strain nr	Species	MERO	COL	AMP	AZI	TAZ	CHL	CIP	FOT	GEN	NAL	SMX	TET	TMP	TGC
EURL EC 9.1	<i>E. coli</i>	0,5	<=1	>64	>64	<=0.5	128	<=0.015	<=0.25	>32	1	>1024	>64	>32	<=0.25
EURL EC 9.2	<i>E. coli</i>	<=0.03	<=1	<=1	4	0.12	8	<=0.015	<=0.06	1	2	<=8	<=2	<=0.25	<=0.25
EURL EC 9.3	<i>E. coli</i>	<=0.03	<=1	>64	8	2	4	<=0.015	64	0.5	2	<=16	<=2	<=0.25	<=0.25
EURL EC 9.4	<i>E. coli</i>	<=0.03	<=1	>64	8	8	>128	0.5	8	1	>128	>1024	>32	>32	<=0.25
EURL EC 9.5	<i>E. coli</i>	<=0.03	<=1	>64	4	8	>128	2	2	1	128	>1024	64	<=0.25	<=0.25
EURL EC 9.6	<i>E. coli</i>	<=0.03	<=1	4	4	0.25	0.5	0.5	<=0.06	1	>128	16	4	<=0.25	<=0.25
EURL EC 9.7	<i>E. coli</i>	0.06	<=1	>64	8	16	8	>8	32	1	>128	>1024	>64	<=0.25	1
EURL EC 9.8	<i>E. coli</i>	<=0,03	<=1	>64	8	4	32	<=0,015	4	1	2	>1024	2	0,5	<=0,25

Strain nr	Species	MERO	COL	AMP	AZI	TAZ	CHL	CIP	FOT	GEN	NAL	SMX	TET	TMP	TGC
EURL EC 9.1	<i>E. coli</i>	R	S	R	R	S	R	S	S	R	S	R	R	R	S
EURL EC 9.2	<i>E. coli</i>	S	S	S	S	S	S	S	S	S	S	S	S	S	S
EURL EC 9.3	<i>E. coli</i>	S	S	R	S	R	S	S	R	S	S	S	S	S	S
EURL EC 9.4	<i>E. coli</i>	S	S	R	S	R	R	R	R	S	R	R	R	R	S
EURL EC 9.5	<i>E. coli</i>	S	S	R	S	R	R	R	R	S	R	R	R	S	S
EURL EC 9.6	<i>E. coli</i>	S	S	S	S	S	S	R	S	S	R	S	S	S	S
EURL EC 9.7	<i>E. coli</i>	S	S	R	S	R	S	R	R	S	R	R	R	S	S
EURL EC 9.8	<i>E. coli</i>	S	S	R	S	R	R	S	R	S	S	R	S	S	S

Panel 2

Strain nr	Species	MERO	FEP	FOX	TAZ	FOT	T/C	F/C	IMI	ETP	TRM	ESBL conclusion
EURL EC 9.1	<i>E. coli</i>	0,5	0,12	4	<=0,25	<=0,25	<=0,12	0,12	1	0,5	128	Presumptive carbapenemase
EURL EC 9.2	<i>E. coli</i>											not ESC
EURL EC 9.3	<i>E. coli</i>	<=0,03	32	4	2	64	<=0,12	<=0,06	0,12	<=0,015	4	Presumptive ESBL
EURL EC 9.4	<i>E. coli</i>	<=0,03	0,25	32	8	8	8	8	0,25	0,03	4	Presumptive AMPC
EURL EC 9.5	<i>E. coli</i>	<=0,03	0,25	4	8	2	<=0,12	<=0,06	<=0,12	<=0,015	4	Presumptive ESBL
EURL EC 9.6	<i>E. coli</i>											not ESC
EURL EC 9.7	<i>E. coli</i>	<=0,03	8	32	16	32	8	4	0,25	0,03	8	Presumptive AMPC+ ESBL
EURL EC 9.8	<i>E. coli</i>	<=0,03	0,5	4	4	4	<=0,12	<=0,06	0,25	<=0,015	8	Presumptive ESBL

Strain nr	Species	MERO	FEP	FOX	TAZ	FOT	T/C	F/C	IMI	ETP	TRM
EURL EC 9.1	<i>E. coli</i>	R	S	S	S	S	NA	NA	R	R	NA
EURL EC 9.2	<i>E. coli</i>										
EURL EC 9.3	<i>E. coli</i>	S	R	S	R	R	NA	NA	S	S	NA
EURL EC 9.4	<i>E. coli</i>	S	R	R	R	R	NA	NA	S	S	NA
EURL EC 9.5	<i>E. coli</i>	S	R	S	R	R	NA	NA	S	S	NA
EURL EC 9.6	<i>E. coli</i>										
EURL EC 9.7	<i>E. coli</i>	S	R	R	R	R	NA	NA	S	S	NA
EURL EC 9.8	<i>E. coli</i>	S	R	S	R	R	NA	NA	S	S	NA

	Resistant
NA	Not applicable or not testet

Abbreviations: AMP- ampicillin, AZI- Azithromycin, , CHL-chloramphenicol, CIP- ciprofloxacin, COL- colistin, ETP- ertapenem, FEP- cefepime, FOT- cefotaxime, FOT/cla- cefotaxime/clav acid, GEN- gentamicin, IMI- imipenem, MER- meropenem, , NAL- nalidixic acid, SMX- sulphamethoxazole, TAZ- ceftazidime, TAZ/CLA- Ceftazidime/clav acid, TET- tetracycline, TMP- trimethoprim, TGC- tigecycline, TRM- temocillin.

G00-06-001/01.12.2014

EURL-AR External Quality Assurance System (EQAS) 2015:-*Escherichia coli*, staphylococci and enterococci

Id: «Lab_no_»

«Name»

«Institute__»

«Country»

Lyngby, 8th June 2015

Dear «Name»

Please find enclosed the bacterial strains for the EURL-AR EQAS 2015. Upon arrival to your laboratory, the strains should be stored dark and at 4°C for stabs, and dark and cool for freeze-dried strains.

On the EURL-AR-website (www.eurl-ar.eu) the following documents relevant for the EURL-AR EQAS are available:

- Protocol for *E. coli*, staphylococci and enterococci including test forms
- Instructions for Opening and Reviving Lyophilised Cultures
- Subculture and Maintenance of Quality Control Strains

We ask you to examine the eight *E. coli*, *enterococci* and *S. aureus* strains that we send to you by performing antimicrobial susceptibility testing. In the protocol you can find detailed description of the procedures to follow. Additionally, you can find a description of the procedure to enter your results into the interactive web database. For accessing the database, you need this username and password:

Your username: «Username»

Your password: «Password»

Please keep this document
Your username and password will not appear in other documents

Results should be entered in the database no later than **4th September 2015**. Please acknowledge receipt of this parcel immediately upon arrival (to licav@food.dtu.dk) and do not hesitate to contact me for further information.

Yours sincerely,

Lina Cavaco



PROTOCOL

For antimicrobial susceptibility testing of *Escherichia coli*, enterococci and staphylococci

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1 INTRODUCTION

The organisation and implementation of an External Quality Assurance System (EQAS) on antimicrobial susceptibility testing (AST) of *E. coli*, enterococci and staphylococci is among the tasks of the EU Reference Laboratory for Antimicrobial Resistance (EURL-AR). The EC/Ent/Staph EQAS 2015 will include AST of eight *E. coli*, eight enterococci and eight staphylococci strains and AST of reference strains *E. coli* ATCC 25922 (CCM 3954), *E. faecalis* ATCC 29212 (CCM 4224), and *S. aureus* ATCC 29213 (CCM 4223).

The above-mentioned reference strains are included in the parcel only for new participants of the EQAS who did not receive them previously. The reference strains are original CERTIFIED cultures provided free of charge, and should be used for future internal quality control for antimicrobial susceptibility testing in your laboratory. The reference strains will not be included in the years to come. Therefore, please take proper care of these strains. Handle and maintain them as suggested in the manual 'Subculture and Maintenance of QC Strains' available on the EURL-AR website (see www.eurl-ar.eu).

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Various aspects of the proficiency test scheme may from time to time be subcontracted. When subcontracting occurs it is placed with a competent subcontractor and the National Food Institute is responsible to the scheme participants for the subcontractor's work.

2 OBJECTIVES

This EQAS aims to support laboratories to assess and, if necessary, to improve the quality of results obtained by AST of pathogens of food- and animal-origin, with special regard to *E. coli*, enterococci and staphylococci. Further objectives are to evaluate and improve the comparability of surveillance data on antimicrobial susceptibility of *E. coli*, enterococci and staphylococci reported to EFSA by different laboratories.

3 OUTLINE OF THE EC/ENT/STAPH EQAS 2015

Shipping, receipt and storage of strains

In June 2015, the National Reference Laboratories for Antimicrobial Resistance (NRL-AR) will receive a parcel containing eight *E. coli*, eight enterococci and eight staphylococci strains from the National Food Institute. This parcel will also contain reference strains, but only for participants who did not receive them previously. All strains belong to UN3373, Biological substance, category B. Extended spectrum beta-lactamase (ESBL)-producing strains as well as carbapenemase-producing strains and methicillin resistant *Staphylococcus aureus* (MRSA) will be included in the selected material.

The reference strains are shipped lyophilised, while the test strains are stab cultures. On arrival, the stab cultures must be subcultured, and all cultures should be adequately stored until testing. A suggested procedure for reconstitution of the lyophilised reference strains is presented below.

Suggested procedure for reconstitution of the lyophilised reference strains

Please refer to the document 'Instructions for opening and reviving lyophilised cultures' reported on the EURL-AR-website (see www.eurl-ar.eu).

Antimicrobial susceptibility testing

The strains should be tested for susceptibility to the antimicrobials listed in Tables 1, 2 and 3, using the method implemented in your laboratory for performing monitoring for EFSA and applying the interpretative criteria listed below.

Participants should perform minimum inhibitory concentration (MIC) determination using the methods stated in the EC regulation EC 652/2013. For staphylococci MIC methods should be used as well, according to the EFSA recommendations and the antimicrobials to test are those stated under the EFSA technical specifications (see Table 3). For interpretation of the results, use the cut-off values listed in Tables 1, 2, 3 and 4 in this document. These values (except where indicated) represent the current epidemiological cut-off values developed by EUCAST (www.eucast.org), and

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allow categorisation of bacterial isolates into two categories: Resistant or susceptible. A categorisation as intermediate is not accepted.

Participants will not be allowed to use disk diffusion as the current regulation and recommendations only focus on MIC testing.

3.1.1 *E. coli*

Table 1: Antimicrobials recommended for AST of *Escherichia coli* and interpretative criteria according to table 1 in EC regulation 652/2013

Antimicrobials for <i>E. coli</i>	MIC (µg/mL) R is >
Ampicillin, AMP	8
Azithromycin, AZI	16*
Cefotaxime, FOT	0.25
Ceftazidime, TAZ	0.5
Chloramphenicol, CHL	16
Ciprofloxacin, CIP	0.06
Colistin, COL	2
Gentamicin, GEN	2
Meropenem, MERO	0.125
Nalidixic acid, NAL	16
Sulfamethoxazole, SMX	64
Tetracycline, TET	8
Tigecycline, TGC	1
Trimethoprim, TMP	2

* Tentative ECOFF established from EFSA data.

Plasmid-mediated quinolone resistance

When performing antimicrobial susceptibility testing of *E. coli*, the interpretative criteria listed in Table 1 for results obtained by MIC-determination should be able to detect plasmid mediated quinolone resistant test strains.

Beta-lactam resistance

Confirmatory tests for ESBL production are mandatory on all strains resistant to cefotaxime (FOT), ceftazidime (TAZ) or meropenem and should be performed by testing the second panel of antimicrobials (Table 2 in this document corresponding to Table 4 in EC regulation 652/2013).

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Table 2: Antimicrobials recommended for additional AST of *Escherichia coli* resistant to cefotaxime, ceftazidime or meropenem and interpretative criteria according to table 4 in EC regulation 652/2013

Antimicrobials for <i>E. coli</i>	MIC (µg/mL) R is >
Cefepime, FEP	0.125
Cefotaxime, FOT	0.25
Cefotaxime + clavulanic acid (F/C)	Not applicable
Cefoxitin, FOX	8
Ceftazidime, TAZ	0.5
Ceftazidime + clavulanic acid (T/C)	Not applicable
Ertapenem, ETP	0.06
Imipenem, IMI	0.5
Meropenem, MERO	0.125
Temocillin, TRM	Not available*

*Where no interpretative criteria are available, we request the participants upload the MIC value obtained, and do not select an interpretation.

Confirmatory test for ESBL production requires use of both cefotaxime (FOT) and ceftazidime (TAZ) alone and in combination with a β -lactamase inhibitor (clavulanic acid). Synergy is defined either as i) a ≥ 3 twofold concentration decrease in an MIC for either antimicrobial agent tested in combination with clavulanic acid vs. its MIC when tested alone (MIC FOT : FOT/CL or TAZ : TAZ/CL ratio ≥ 8) (CLSI M100 Table 2A; Enterobacteriaceae). The presence of synergy indicates ESBL production. Resistance to cefepime gives further indication of ESBL production, but is not essential.

Confirmatory test for carbapenemase production requires the testing of meropenem (MERO).

Detection of AmpC-type beta-lactamases can be performed by testing the bacterium for susceptibility to cefoxitin (FOX). Resistance to FOX could indicate the presence of an AmpC-type beta-lactamase, that may be verified by PCR and sequencing.

The classification of the phenotypic results should be based on the most recent EFSA recommendations (EFSA 2012), indicating the strains as:

- Presumptive ESBL: strains with positive synergy test, susceptible to cefoxitin and resistant to cefepime
- Presumptive ESBL+pAmpC: strains with positive or negative synergy test, resistant to cefoxitin and resistant to cefepime
- Presumptive pAmpC phenotype: strains with negative synergy test, resistant to cefoxitin and susceptible to cefepime
- Presumptive carbapenemase phenotype: strain resistant to meropenem
- Unusual phenotype: any other combinations

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(However we recommend that strains which show synergy with clavulanic acid for at least one of the third generation cephalosporins, cefotaxime or ceftazidime, should be considered ESBL, independently of the cefepime result)

3.1.2 Enterococci

Table 3: Antimicrobials recommended for AST of *Enterococcus* spp. and interpretative criteria according to table 3 in EC regulation 652/2013.

Antimicrobials for enterococci	MIC (µg/mL) R is > <i>E. faecium</i>	MIC (µg/mL) R is > <i>E. faecalis</i>
Ampicillin, AMP	4	4
Chloramphenicol, CHL	32	32
Ciprofloxacin, CIP	4	4
Daptomycin, DAP	4	4
Erythromycin, ERY	4	4
Gentamicin, GEN	32	32
Linezolid, LZD	4	4
Quinupristin-dalfopristin (Synercid), SYN	4*	Not applicable
Teicoplanin, TEI	2	2
Tetracycline, TET	4	4
Tigecycline, TGC	0.25	0.25
Vancomycin, VAN	4	4

*DANMAP 2009 (www.danmap.org)

Identification of the *Enterococcus* spp.

Species identification of the Enterococci must be performed by the NRLs using in-house methods or adopting the protocol available on the EURL-AR website under: www.eurl-ar.eu/233-protocols.htm.

3.1.3 Staphylococci

Eight staphylococci strains will be sent to be tested in the AST component of the EQAS 2015.

Identification of MRSA

Confirmation of *mecA* and/or *mecC* presence is mandatory in this EQAS. For this purpose, you are recommended to use the PCR method protocol recommended by the EURL-AR (www.eurl-ar.eu/233-protocols.htm) and upload the result as 'positive' or 'negative'. According to CLSI recommendations (M100, Table 2C), all MRSA should be regarded as resistant to all β -lactam antibiotics.



Table 4: Antimicrobials recommended for AST of *Staphylococcus aureus* and interpretative criteria according to EFSA technical specifications (EFSA 2012)

Antimicrobials for <i>S. aureus</i>	MIC (µg/mL) R is >
Cefoxitin, FOX	4
Chloramphenicol, CHL	16
Ciprofloxacin, CIP	1
Clindamycin, CLN	0.25
Erythromycin, ERY	1
Gentamicin, GEN	2
Linezolid, LZD	4
Mupirocin, MUP	1
Quinupristin-dalfopristin (Synercid), SYN	1
Sulfamethoxazole, SMX	128
Sulfamethoxazole+Trimethoprim, SXT	0.5
Tetracycline, TET	1
Tiamulin, TIA	2
Trimethoprim, TMP	2
Vancomycin, VAN	2

*CLSI M100 Table 2C

4 REPORTING OF RESULTS AND EVALUATION

Please write your results in the test forms, and enter your results into the interactive web database. In addition, we kindly ask you to report in the database the tested MIC range for the staphylococci tests, (for this organism, only, as it is not covered by the EC regulation on MIC testing). Finally, if **you did not use the cut-off values recommended in the protocol for interpretation of *Staphylococcus* AST results**, please report the breakpoints used in the database.

4.1 General recommendations for data upload

We recommend reading carefully the description reported in paragraph 5 before entering your results in the web database. **Results must be submitted no later than September 4th 2015.** After the deadline when all participants have uploaded results, you will be able to login to the database once again, and to view and print an automatically generated report evaluating your results. Results in agreement with the expected interpretation are categorised as ‘correct’, while results deviating from the expected interpretation are categorised as ‘incorrect’.

If you experience difficulties in entering your results, please contact us directly.

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All results will be summarized in a report which will be publicly available. The data in the report will be presented with laboratory codes. A laboratory code is known to the individual laboratory, whereas the complete list of laboratories and their codes is confidential and known only to the EURL-AR and the EU Commission. All conclusions will be public.

If you have questions, please do not hesitate to contact the EQAS Coordinator:

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National Food Institute
Technical University of Denmark
Søltofts Plads 221, DK-2800 Lyngby

Denmark

Tel: +45 3588 6601

Fax: +45 3588 6341

E-mail: suska@food.dtu.dk

5 HOW TO ENTER RESULTS IN THE INTERACTIVE DATABASE

Please read carefully this paragraph before entering the web page.

Remember that you need by your side the completed test forms and the breakpoint values you used.

Enter the EURL-AR EQAS 2015 start web page (<http://eurl-ar.food.dtu.dk>), write your username and password in lower-cases and press enter. Your username and password are indicated in the letter following your strains. Do not hesitate to contact us if you experience problems with the login.

You can browse back and forth by using the Home or back keys, but please remember to save your inputs before.

5.1 AST of *E. coli*, enterococci and staphylococci

Click on either "*E. coli*", "enterococci" or "staphylococci" for input of test results based on the results you are going to upload.

Click on "Start of Data Entry - Methods and Breakpoints"

In the next page, you can navigate among fields with the Tab-key and the mouse.

Complete the fields related to the method used for antimicrobial susceptibility testing and the brand of MIC trays, etc.

Click on "save" and then go back using the tab "home" and enter another test page to upload results

EU Reference Laboratory for Antimicrobial Resistance

External Quality Assurance System (EQAS) 2015



In the data entry pages, enter the obtained values and the interpretation (R, resistant or S, susceptible) for each *E. coli*, enterococcus and staphylococcus strain.

For *E. coli* strains, remember to report also the results for the ESBL detection tests.

For *S. aureus* strains, remember to report also the results for presence/absence of methicillin resistance.

If you did not test for susceptibility to a given antimicrobial, please leave the field empty.

Click on “save“ and then go back using the tab “home” and enter another test page to upload results.

When uploading data on the reference strains, please enter MIC values in µg/ml. Remember to use the operator keys to show symbols like “equal to”, etc.

Click on “save“.

Review the input pages by browsing through the pages and make corrections if necessary.

Remember to save a page if you make corrections. If you press home a page without saving changes, you will see an error screen. In this case, click on “save“ to save your results, browse back to the page and then continue.

Please complete the evaluation form.

Before approving your input, please be sure that you have filled in all the relevant fields because **YOU CAN ONLY APPROVE ONCE!** The approval blocks your data entry in the interactive database.

**EU Reference Laboratory for Antimicrobial Resistance
External Quality Assurance System (EQAS) 2015**



Antimicrobial susceptibility testing of *Escherichia coli*, enterococci
and staphylococci

TEST FORMS

Name:

Name of laboratory:

Name of institute:

City:

Country:

E-mail:

Fax:

Comments:



TEST FORMS METHODS - Enterococci

Which method did you use for antimicrobial susceptibility testing of enterococci in this EQAS:

- ☐ MIC – Microtitre
☐ MIC – Agar dilution

Brand:

How many *Enterococcus* spp. isolates does your laboratory annually isolate:

How many *Enterococcus* spp. isolates does your laboratory annually test for antimicrobial susceptibility by a MIC method:

Which method was followed for the preparation of the inoculum (please describe)

- Which standard was followed (TREK, CLSI...)
- Which solvent was used for the preparation of the 0.5 McFarland solution (water, saline)
- Please describe in detail how you prepared the dilution of the inoculum (including the volume in final MH-dilution and intended dilution level; e.g. diluted 1:1000 by adding 10µl of 0.5 McFarland solution in 10ml MH broth, for an expected inoculum of 1×10^5 CFU/ml)

Comments or additional information:



TEST FORMS METHODS - staphylococci

Which method did you use for antimicrobial susceptibility testing of staphylococci in this EQAS:

- ☐ MIC – Microtitre
☐ MIC – Agar dilution

Brand:

How many *Staphylococcus* spp. isolates does your laboratory annually isolate:

How many *Staphylococcus* spp. isolates does your laboratory annually test for antimicrobial susceptibility by a MIC method:

Which method was followed for the preparation of the inoculum (please describe)

- Which standard was followed (TREK, CLSI...)
- Which solvent was used for the preparation of the 0.5 McFarland solution (water, saline)
- Please describe in detail how you prepared the dilution of the inoculum (including the volume in final MH-dilution and intended dilution level; e.g. diluted 1:1000 by adding 10µl of 0.5 McFarland solution in 10ml MH broth, for an expected inoculum of 1×10^5 CFU/ml)

Comments or additional information:

Antimicrobial	General info			
	The relevant information in the two columns below should be reported			
	Test-range for MIC (µg/mL)	Resistant (µg/mL)	Intermediate (µg/mL)	Susceptible (µg/mL)
Cefoxitin, FOX		≤		≥
Chloramphenicol, CHL		≤		≥
Ciprofloxacin, CIP		≤		≥
Clindamycin, CLN		≤		≥
Erythromycin, ERY		≤		≥
Gentamicin, GEN		≤		≥
Linezolid, LZD		≤		≥
Mupirocin, MUP		≤		≥
Quin.-Dalf. (Synercid), SYN		≤		≥
Sulfamethoxazole, SMX		≤		≥
Sulfamethoxazole + Trimethoprim SXT		≤		≥
Tetracycline, TET		≤		≥
Tiamulin, (TIA)		≤		≥
Trimethoprim, TMP		≤		≥
Vancomycin, VAN		≤		≥



TEST FORMS METHODS – *E. coli*

Which method did you use for antimicrobial susceptibility testing of *E. coli* in this EQAS:

- ☐ MIC – Microtitre
☐ MIC – Agar dilution

Brand:

Incubation conditions: °C/ h

How many *E. coli* isolates does your laboratory annually isolate:

How many *E. coli* isolates does your laboratory annually test for antimicrobial susceptibility by a MIC method:

Which method was followed for the preparation of the inoculum (please describe)

- Which standard was followed (TREK, CLSI...)
- Which solvent was used for the preparation of the 0.5 McFarland solution (water, saline)
- Please describe in detail how you prepared the dilution of the inoculum (including the volume in final MH-dilution and intended dilution level; e.g. diluted 1:1000 by adding 10µl of 0.5 McFarland solution in 10ml MH broth, for an expected inoculum of 1×10^5 CFU/ml)

Comments or additional information:



TEST FORM - Enterococci

Strain	Antimicrobial	Results and interpretation		
		≤ >	MIC-value (µg/ml)	S / R
Enterococci EURL ENT. 9.X <input type="checkbox"/> <i>E. faecium</i> <input type="checkbox"/> <i>E. faecalis</i>	Ampicillin AMP			
	Chloramphenicol, CHL			
	Ciprofloxacin, CIP			
	Daptomycin, DAP			
	Erythromycin, ERY			
	Gentamicin, GEN			
	Linezolid, LZD			
	Quin.-Dalf. (Synercid), SYN			
	Teicoplanin, TEI			
	Tetracycline, TET			
	Tigecycline, TGC			
	Vancomycin, VAN			

Strain	Antimicrobial	Results and interpretation		
		≤ >	MIC-value (µg/ml)	S / R
Enterococci EURL ENT. 9.X <input type="checkbox"/> <i>E. faecium</i> <input type="checkbox"/> <i>E. faecalis</i>	Ampicillin AMP			
	Chloramphenicol, CHL			
	Ciprofloxacin, CIP			
	Daptomycin, DAP			
	Erythromycin, ERY			
	Gentamicin, GEN			
	Linezolid, LZD			
	Quin.-Dalf. (Synercid), SYN			
	Teicoplanin, TEI			
	Tetracycline, TET			
	Tigecycline, TGC			
	Vancomycin, VAN			



TEST FORM - Enterococci

Antimicrobial susceptibility testing of reference strain *Enterococcus faecalis* ATCC 29212

Antimicrobial	MIC-value (µg/ml)
Ampicillin, AMP	
Chloramphenicol, CHL	
Ciprofloxacin, CIP	
Daptomycin, DAP	
Erythromycin, ERY	
Gentamicin, GEN	
Linezolid, LZD	
Quinupristin-Dalfopristin (Synercid), SYN	
Teicoplanin, TEI	
Tetracycline, TET	
Tigecycline, TIG	
Vancomycin, VAN	



TEST FORMS - Staphylococci

Strain	Antimicrobial	Results and interpretation		
		≤ >	MIC-value (µg/ml)	S / R
<i>S. aureus</i> EURL ST 9.X	Cefoxitin, FOX			
	Chloramphenicol, CHL			
	Ciprofloxacin, CIP			
	Clindamycin, CLN			
	Erythromycin, ERY			
	Gentamicin, GEN			
	Linezolid, LZD			
	Mupirocin, MUP			
	Quino-dalfopristin (Synercid), SYN			
	Sulfamethoxazole, SMX			
	Sulfamethoxazole+Trimethoprim, SXT			
	Tetracycline, TET			
	Tiamulin, TIA			
	Trimethoprim, TMP			
	Vancomycin, VAN			

Methicillin resistance (MRSA)	<input type="checkbox"/> Positive	<input type="checkbox"/> Negative
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TEST FORM - Staphylococci

Antimicrobial susceptibility testing of reference strain *S. aureus* ATCC 29213 (MIC)

Antimicrobial	MIC-value (µg/ml)
Cefoxitin, FOX	
Chloramphenicol, CHL	
Ciprofloxacin, CIP	
Clindamycin, CLN	
Erythromycin, ERY	
Gentamicin, GEN	
Linezolid, LZD	
Mupirocin, MUP	
Quino-dalfo (Synercid), SYN	
Sulfamethoxazole, SMX	
Sulfamethoxazole+ Trimethoprim , SXT	
Tetracycline, TET	
Tiamulin, TIA	
Trimethoprim, TMP	
Vancomycin, VAN	



TEST FORM – *E. coli*

Strain	Antimicrobial	Results and interpretation		
		≤ >	MIC-value (µg/ml)	S / R
<i>E. coli</i> EURL EC 9.X	Ampicillin, AMP			
	Azithromycin, AZT			
	Cefotaxime, FOT			
	Ceftazidime, TAZ			
	Chloramphenicol, CHL			
	Ciprofloxacin CIP			
	Colistin, COL			
	Gentamicin, GEN			
	Meropenem, MERO			
	Nalidixic acid, NAL			
	Sulfamethoxazole, SMX			
	Tetracycline, TET			
	Tigecycline, TGC			
	Trimethoprim, TMP			

All strains resistant to cefotaxime (FOT), ceftazidime (TAZ) or meropenem (MERO) should be included for testing in the second panel confirmatory tests for ESBL or carbapenemase production. See further description of confirmatory tests in the protocol section '3.1.1E. coli'.

Strain	Antimicrobial	Results and interpretation		
		≤ >	MIC-value (µg/ml)	S / R
<i>E. coli</i> EURL EC 9.X	Cefepime, FEP			
	Cefotaxime, FOT			
	Cefotaxime + clavulanic acid (F/C)			
	Cefoxitin, FOX			
	Ceftazidime, TAZ			
	Ceftazidime+ clavulanic acid (T/C)			
	Ertapenem, ETP			
	Imipenem, IMI			
	Meropenem, MERO			
	Temocillin, TRM			

☐ Presumptive ESBL
 ☐ Presumptive pAmpC
 ☐ Unusual phenotype

☐ Presumptive ESBL+ pAmpC
 ☐ Presumptive carbapenemase
 ☐ No ESBL, AmpC- or carbapenemase

Comments (include optional genotype or other results):



TEST FORM – *E. coli*

Antimicrobial susceptibility testing of reference strain *E. coli* ATCC 25922

	Antimicrobial	MIC-value (µg/ml)
1 st panel	Ampicillin, AMP	
	Azithromycin, AZT	
	Cefotaxime, FOT	
	Ceftazidime, TAZ	
	Chloramphenicol, CHL	
	Ciprofloxacin, CIP	
	Colistin, COL	
	Gentamicin, GEN	
	Meropenem, MERO	
	Nalidixic acid, NAL	
	Sulfamethoxazole, SMX	
	Tetracycline, TET	
	Tigecycline, TGC	
	Trimethoprim, TMP	
2 nd panel	Cefepime, FEP	
	Cefotaxime, FOT	
	Cefotaxime, + clavulanic acid (F/C)	
	Cefoxitin, FOX	
	Ceftazidime, TAZ	
	Ceftazidime + clavulanic acid (T/C)	
	Ertapenem, ETP	
	Imipenem, IMI	
	Meropenem, MERO	
	Temocillin, TRM	

INSTRUCTIONS FOR OPENING AND REVIVING LYOPHILISED CULTURES

Instructions adjusted from Czech Collection of Microorganisms (CCM) document 'Instructions for Opening and Reviving of Freeze-Dried Bacteria and Fungi' available on <http://www.sci.muni.cz>.

Lyophilised cultures are supplied in vacuum-sealed ampoules. Care should be taken in opening the ampoule. All instructions given below should be followed closely to ensure the safety of the person who opens the ampoule and to prevent contamination of the culture.

- a. Check the number of the culture on the label inside the ampoule
- b. Make a file cut on the ampoule near the middle of the plug (see Figure 1)
- c. Disinfect the ampoule with alcohol-dampened gauze or alcohol-dampened cotton wool from just below the plug to the pointed end
- d. Apply a red-hot glass rod to the file cut to crack the glass and allow air to enter slowly into the ampoule
- e. Remove the pointed end of the ampoule into disinfectant
- f. Add about 0.3 ml appropriate broth to the dried suspension using a sterile Pasteur pipette and mix carefully to avoid creating aerosols. Transfer the contents to one or more suitable solid and /or liquid media
- g. Incubate the inoculated medium at appropriate conditions for several days
- h. Autoclave or disinfect effectively the used Pasteur pipette, the plug and all the remains of the original ampoule before discarding

Notes:

- Cultures should be grown on media and under conditions as recommended in the CCM catalogue (see <http://www.sci.muni.cz>)
- Cultures may need at least one subculturing before they can be optimally used in experiments
- Unopened ampoules should be kept in a dark and cool place!

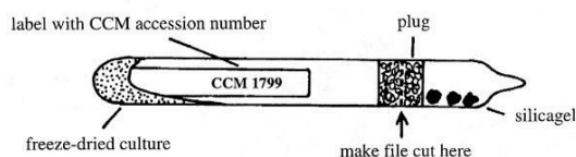


Figure 1: from CCM document 'Instructions for Opening and Reviving of Freeze-Dried Bacteria and Fungi' available on <http://www.sci.muni.cz>

SUBCULTURE AND MAINTENANCE OF QUALITY CONTROL STRAINS

1.1 Purpose

Improper storage and repeated subculturing of bacteria can produce alterations in antimicrobial susceptibility test results. The Clinical and Laboratory Standards Institute (CLSI, formerly NCCLS) has published a guideline for Quality Control (QC) stock culture maintenance to ensure consistent antimicrobial susceptibility test results.

1.2 References

M100-S24, January 2014 (Performance Standards for Antimicrobial Susceptibility Testing)

M7-A9, January 2012 (Methods for Dilution Antimicrobial Susceptibility Test for Bacteria That Grow Aerobically; Approved Standard)

1.3 Definition of Terms

Reference Culture: A reference culture is a microorganism preparation that is acquired from a culture type collection.

Reference Stock Culture: A reference stock culture is a microorganism preparation that is derived from a reference culture. Guidelines and standards outline how reference stock cultures must be processed and stored.

Working Stock Cultures: A working stock culture is growth derived from a reference stock culture. Guidelines and standards outline how working stock cultures must be processed and how often they can be subcultured.

Subcultures (Passages): A subculture is simply the transfer of established microorganism growth on media to fresh media. The subsequent growth on the fresh media constitutes a subculture or passage. Growing a reference culture or reference stock culture from its preserved status (frozen or lyophilized) is not a subculture. The preserved microorganism is not in a stage of established growth until it is thawed or hydrated and grown for the first time

1.4 Important Considerations

- Do not use disc diffusion strains for MIC determination.
- Obtain QC strains from a reliable source such as ATCC
- CLSI requires that QC be performed either on the same day or weekly (only after 30 day QC validation)
- Any changes in materials or procedure must be validated with QC before implemented
- For example: Agar and broth methods may give different QC ranges for drugs such as glycopeptides, aminoglycosides and macrolides
- Periodically perform colony counts to check the inoculum preparation procedure

- Ideally, test values should be in the middle of the acceptable range
- Graphing QC data points over time can help identify changes in data helpful for troubleshooting problems

1.5 Storage of Reference Strains

Preparation of stock cultures

- Use a suitable stabilizer such as 50% fetal calf serum in broth, 10-15% glycerol in tryptic soy broth, defibrinated sheep blood or skim milk to prepare multiple aliquots.
- Store at -20°C, -70°C or liquid nitrogen. (Alternatively, freeze dry.)
- Before using rejuvenated strains for QC, subculture to check for purity and viability.

Working cultures

- Set up on agar slants with appropriate medium, store at 4-8°C and subculture weekly.
- Replace the working strain with a stock culture at least monthly.
- If a change in the organisms inherent susceptibility occurs, obtain a fresh stock culture or a new strain from a reference culture collection e.g. ATCC.

1.6 Frequency of Testing

Weekly vs. daily testing

Weekly testing is possible if the lab can demonstrate satisfactory performance with daily testing as follows:

- Documentation showing reference strain results from 30 consecutive test days were within the acceptable range.
- For each antimicrobial/organism combination, no more than 3 out of 30 MIC values may be outside the acceptable range.

When the above are fulfilled, each quality control strain may be tested once a week and whenever any reagent component is changed.

Corrective Actions

If an MIC is outside the range in weekly testing, corrective action is required as follows:

- Repeat the test if there is an obvious error e.g. wrong strain or incubation conditions used
- If there is no obvious error, return to daily control testing

The problem is considered resolved only after the reference strain is tested for 5 consecutive days and each drug/organism result is within specification on each day.

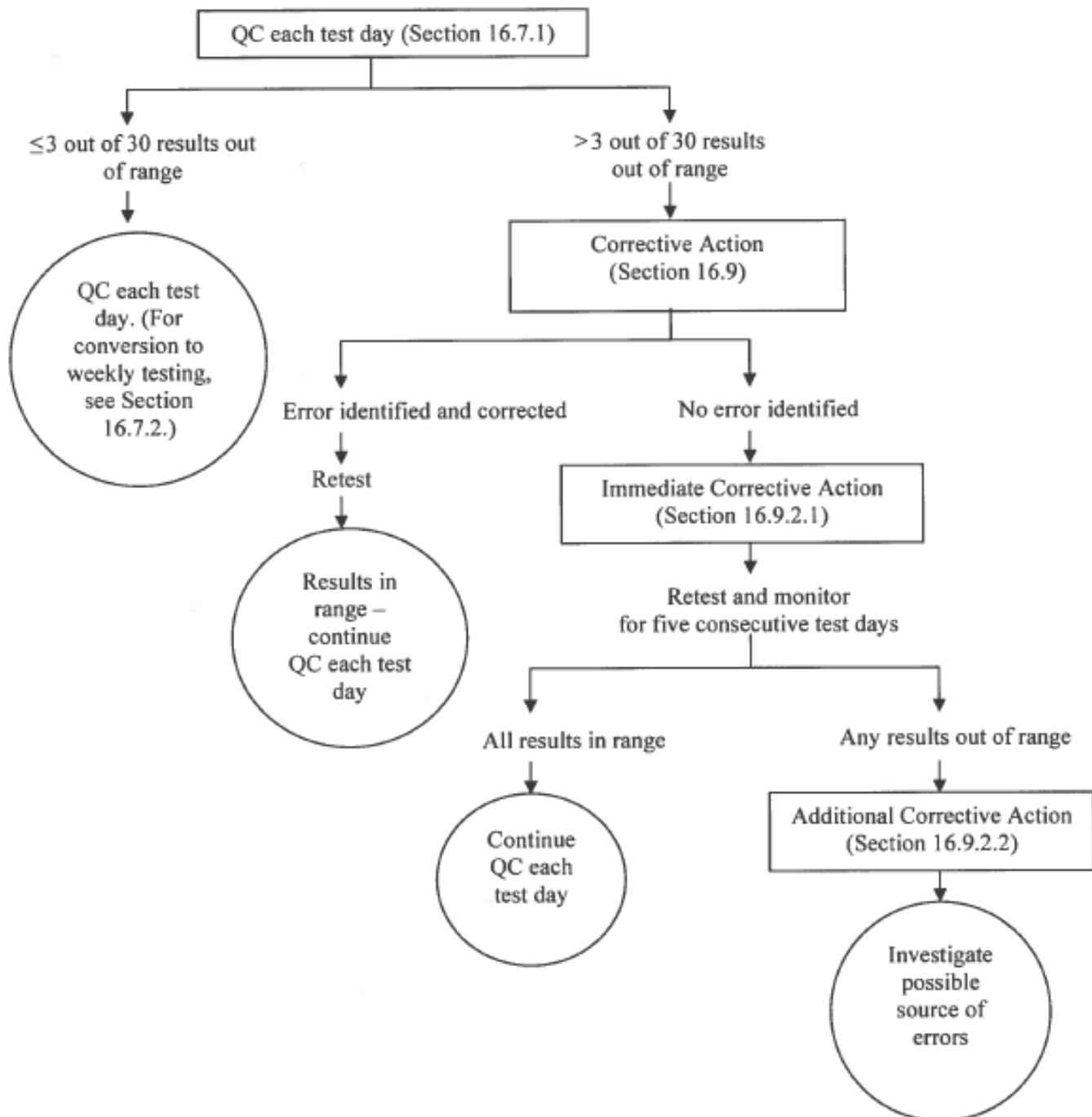
If the problem cannot be resolved, continue daily testing until the errors are identified.

Repeat the 30 days validation before resuming weekly testing.

DAILY MIC QC CHART

Appendix A. Quality Control Protocol Flow Charts

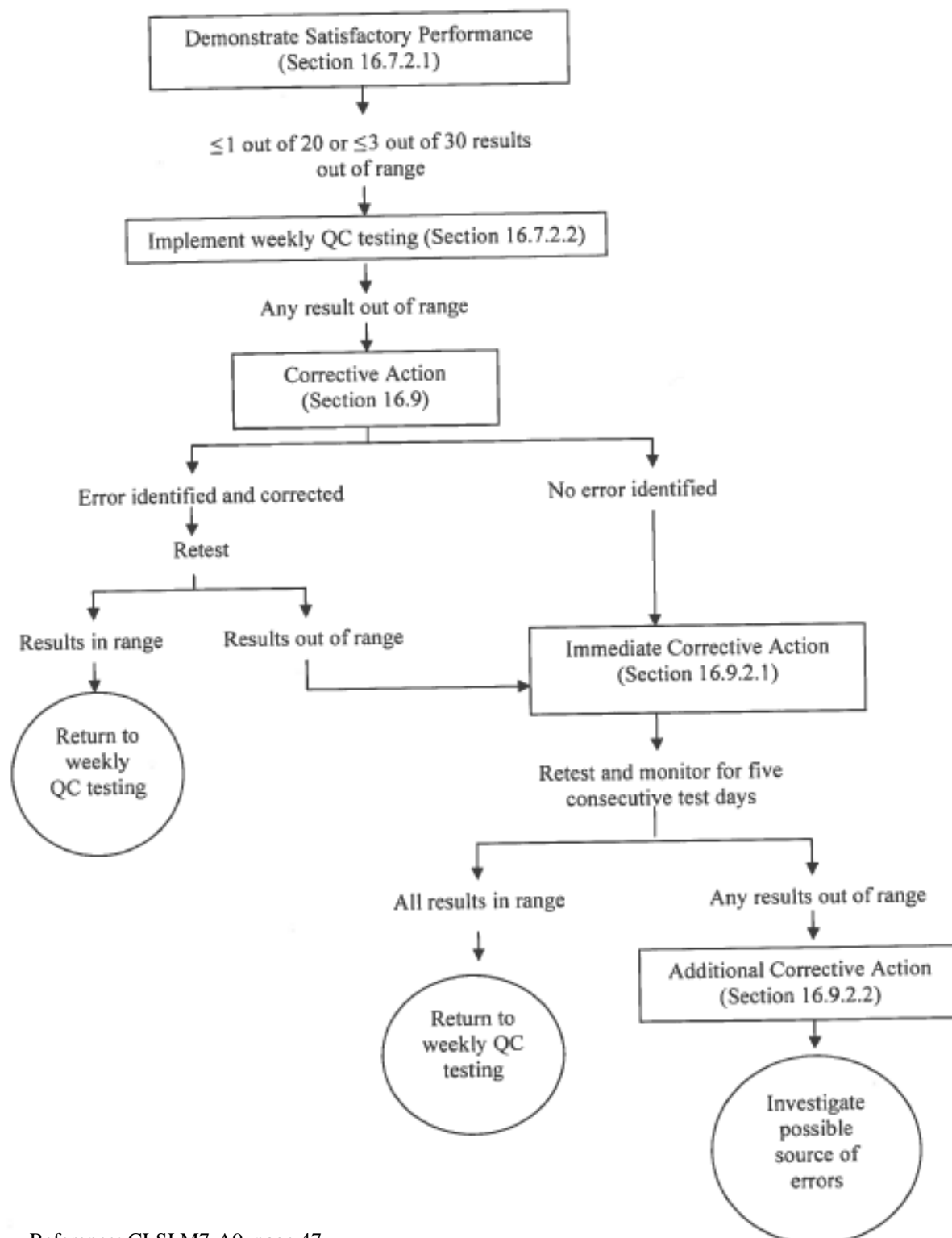
Quality Control (QC) Protocol: Daily Testing



Reference: CLSI M7-A9, page 46

Appendix A. (Continued)

QC Protocol: Weekly Testing



Reference: CLSI M7-A9, page 47

Appendix 5- Quality control ranges for ATCC QC strains*Escherichia coli* ATCC 25922

Panel	Antimicrobial		Min.	Max
1	Ampicillin	AMP	2	8
	Azithromycin	AZI		
	Cefotaxime	FOT	0.03	0.12
	Ceftazidime	TAZ	0.06	0.5
	Chloramphenicol	CHL	2	8
	Ciprofloxacin	CIP	0.004	0.015
	Colistin	COL	0.25	2
	Gentamicin	GEN	0.25	1
	Meropenem	MER	0.008	0.06
	Nalidixic acid	NAL	1	4
	Sulfamethoxazole	SMX	8	32
	Tetracycline	TET	0.5	2
	Tigecycline	TGC	0.03	0.25
	Trimethoprim	TMP	0.5	2
2	Cefepime	FEP	0.015	0.12
	Cefotaxime/clavulanic acid	F/C		
	Cefotaxime	FOT	0.03	0.12
	Cefoxitin	FOX	2	8
	Ceftazidime	TAZ	0.06	0.5
	Ceftazidime/clavulanic acid	T/C		
	Ertapenem	ETP	0.004	0.015
	Imipenem	IMI	0.06	0.25
	Meropenem	MER	0.008	0.06
	Temocillin	TRM		

Enterococcus faecalis ATCC 29212

Antimicrobial		min	max
Daptomycin	DAP	1	4
Linezolid	LZD	1	4
Chloramphenicol	CHL	4	16
Ciprofloxacin	CIP	0.25	2
Gentamicin	GEN	4	16
Erythromycin	ERY	1	4
Teicoplanin	TEI	0.25	1
Tetracycline	TET	8	32
Tigecycline	TGC	0.03	0.12
Vancomycin	VAN	1	4
Ampicillin	AMP	0.5	2
Quinopristin_Dalfo	SYN	2	8

Staphylococcus aureus ATCC 29213

Antimicrobial		min	max
Cefoxitin	FOX	1	4
Chloramphenicol	CHL	2	16
Ciprofloxacin	CIP	0.12	0.5
Clindamycin	CLN	0.06	0.25
Erythromycin	ERY	0.25	1
Gentamicin	GEN	0.12	1
Linezolid	LZD	1	4
Mupirocin	MUP		
Quinopristin_Dalfo	SYN	0.25	1
Sulfamethoxazole	SMX	32	128
Sulfamethoxazole-Trimethoprim	SXT	0	0.5
Tetracycline	TET	0.12	1
Tiamulin	TIA		
Trimethoprim	TMP	1	4
Vancomycin	VAN	0.5	2

Appendix 6a- Test results from reference strain *Enterococcus faecalis* ATCC 29212

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
2	Ampicillin	=	1	0,5	2	1
2	Chloramphenicol	=	8	4	16	1
2	Ciprofloxacin	=	0,5	0,25	2	1
2	Daptomycin	=	2	1	4	1
2	Erythromycin	<=	1	1	4	1
2	Linezolid	=	2	1	4	1
2	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
2	Teicoplanin	<=	0,5	0,25	1	1
2	Tetracycline	=	32	8	32	1
2	Tigecycline	=	0,5	0,03	0,125	0
2	Vancomycin	=	4	1	4	1
2	Gentamicin	<=	8	4	16	1
9	Ampicillin	=	1	0,5	2	1
9	Ciprofloxacin	=	0,5	0,25	2	1
9	Daptomycin	=	2	1	4	1
9	Erythromycin	=	2	1	4	1
9	Gentamicin	<=	8	4	16	1
9	Linezolid	=	2	1	4	1
9	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
9	Tetracycline	=	16	8	32	1
9	Tigecycline	=	0,06	0,03	0,125	1
9	Vancomycin	=	2	1	4	1
9	Chloramphenicol	=	8	4	16	1
9	Teicoplanin	<=	0,5	0,25	1	1
11	Ampicillin	<=	0,5	0,5	2	1
11	Chloramphenicol	<=	4	4	16	1
11	Ciprofloxacin	=	1	0,25	2	1
11	Daptomycin	=	2	1	4	1
11	Erythromycin	=	2	1	4	1
11	Linezolid	=	1	1	4	1
11	Quinopristin-dalfopristin (Synercid)	=	2	2	8	1
11	Tetracycline	=	8	8	32	1
11	Tigecycline	<=	0,03	0,03	0,125	1
11	Vancomycin	=	2	1	4	1
11	Gentamicin	<=	8	4	16	1
12	Ampicillin	=	2	0,5	2	1
12	Gentamicin	=	8	4	16	1
12	Linezolid	=	2	1	4	1
12	Tetracycline	=	32	8	32	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
12	Chloramphenicol	=	4	4	16	1
12	Vancomycin	=	2	1	4	1
12	Erythromycin	=	4	1	4	1
16	Ampicillin	=	2	0,5	2	1
16	Chloramphenicol	=	8	4	16	1
16	Ciprofloxacin	=	1	0,25	2	1
16	Daptomycin	=	2	1	4	1
16	Linezolid	=	2	1	4	1
16	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
16	Teicoplanin	<=	0,5	0,25	1	1
16	Tetracycline	=	32	8	32	1
16	Tigecycline	=	0,12	0,03	0,125	1
16	Vancomycin	=	4	1	4	1
16	Gentamicin	=	16	4	16	1
16	Erythromycin	=	2	1	4	1
17	Ampicillin	=	1	0,5	2	1
17	Daptomycin	=	2	1	4	1
17	Erythromycin	=	2	1	4	1
17	Gentamicin	<=	8	4	16	1
17	Linezolid	=	2	1	4	1
17	Quinopristin-dalfopristin (Synercid)	=	4	2	8	1
17	Teicoplanin	<=	0,5	0,25	1	1
17	Tigecycline	=	0,12	0,03	0,125	1
17	Vancomycin	=	2	1	4	1
17	Chloramphenicol	=	8	4	16	1
17	Tetracycline	=	32	8	32	1
17	Ciprofloxacin	=	0,5	0,25	2	1
19	Chloramphenicol	=	8	4	16	1
19	Ciprofloxacin	=	0,5	0,25	2	1
19	Daptomycin	=	2	1	4	1
19	Erythromycin	=	2	1	4	1
19	Linezolid	=	2	1	4	1
19	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
19	Teicoplanin	<=	0,5	0,25	1	1
19	Tigecycline	=	0,25	0,03	0,125	0
19	Vancomycin	=	2	1	4	1
19	Gentamicin	=	16	4	16	1
19	Ampicillin	=	1	0,5	2	1
19	Tetracycline	=	32	8	32	1
20	Ampicillin	=	1	0,5	2	1
20	Chloramphenicol	=	8	4	16	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
20	Ciprofloxacin	=	1	0,25	2	1
20	Erythromycin	=	2	1	4	1
20	Gentamicin	=	16	4	16	1
20	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
20	Teicoplanin	<=	0,5	0,25	1	1
20	Tigecycline	=	0,12	0,03	0,125	1
20	Vancomycin	=	4	1	4	1
20	Daptomycin	=	4	1	4	1
20	Tetracycline	=	32	8	32	1
20	Linezolid	=	2	1	4	1
21	Chloramphenicol	=	4	4	16	1
21	Ciprofloxacin	=	0,5	0,25	2	1
21	Erythromycin	=	1	1	4	1
21	Linezolid	=	1	1	4	1
21	Quinopristin-dalfopristin (Synercid)	>	4	2	8	1
21	Vancomycin	=	1	1	4	1
21	Tetracycline	>	16	8	32	1
21	Gentamicin	=	8	4	16	1
22	Ampicillin	=	1	0,5	2	1
22	Chloramphenicol	=	8	4	16	1
22	Ciprofloxacin	=	0,5	0,25	2	1
22	Daptomycin	=	1	1	4	1
22	Gentamicin	=	16	4	16	1
22	Linezolid	=	2	1	4	1
22	Teicoplanin	<=	0,5	0,25	1	1
22	Tetracycline	=	32	8	32	1
22	Vancomycin	=	2	1	4	1
22	Erythromycin	=	2	1	4	1
23	Ampicillin	=	1	0,5	2	1
23	Chloramphenicol	=	8	4	16	1
23	Ciprofloxacin	=	0,25	0,25	2	1
23	Erythromycin	=	2	1	4	1
23	Gentamicin	<=	8	4	16	1
23	Linezolid	=	1	1	4	1
23	Quinopristin-dalfopristin (Synercid)	=	2	2	8	1
23	Teicoplanin	<=	0,5	0,25	1	1
23	Tigecycline	=	0,06	0,03	0,125	1
23	Vancomycin	=	2	1	4	1
23	Tetracycline	=	8	8	32	1
23	Daptomycin	=	1	1	4	1
25	Ampicillin	=	1	0,5	2	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
25	Chloramphenicol	=	8	4	16	1
25	Ciprofloxacin	=	1	0,25	2	1
25	Erythromycin	=	2	1	4	1
25	Gentamicin	=	16	4	16	1
25	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
25	Teicoplanin	<=	0,5	0,25	1	1
25	Tigecycline	=	0,12	0,03	0,125	1
25	Vancomycin	=	4	1	4	1
25	Daptomycin	=	2	1	4	1
25	Tetracycline	=	32	8	32	1
25	Linezolid	=	2	1	4	1
26	Ampicillin	<=	0,5	0,5	2	1
26	Chloramphenicol	=	8	4	16	1
26	Ciprofloxacin	=	1	0,25	2	1
26	Erythromycin	=	2	1	4	1
26	Gentamicin	<=	8	4	16	1
26	Teicoplanin	<=	0,5	0,25	1	1
26	Tetracycline	=	16	8	32	1
26	Tigecycline	=	0,12	0,03	0,125	1
26	Vancomycin	=	4	1	4	1
26	Linezolid	=	2	1	4	1
26	Daptomycin	=	1	1	4	1
29	Ampicillin	=	1	0,5	2	1
29	Ciprofloxacin	=	0,5	0,25	2	1
29	Daptomycin	=	1	1	4	1
29	Erythromycin	=	4	1	4	1
29	Gentamicin	=	8	4	16	1
29	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
29	Teicoplanin	<=	0,5	0,25	1	1
29	Tetracycline	=	32	8	32	1
29	Vancomycin	=	2	1	4	1
29	Linezolid	=	2	1	4	1
29	Chloramphenicol	=	8	4	16	1
29	Tigecycline	=	0,12	0,03	0,125	1
30	Ampicillin	<=	0,5	0,5	2	1
30	Chloramphenicol	<=	4	4	16	1
30	Ciprofloxacin	=	1	0,25	2	1
30	Erythromycin	=	2	1	4	1
30	Gentamicin	<=	8	4	16	1
30	Linezolid	=	2	1	4	1
30	Quinopristin-dalfopristin (Synercid)	=	4	2	8	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
30	Tigecycline	=	0,12	0,03	0,125	1
30	Vancomycin	=	2	1	4	1
30	Daptomycin	=	2	1	4	1
30	Tetracycline	=	32	8	32	1
30	Teicoplanin	<=	0,5	0,25	1	1
33	Ampicillin	=	1	0,5	2	1
33	Chloramphenicol	=	4	4	16	1
33	Erythromycin	=	4	1	4	1
33	Gentamicin	=	8	4	16	1
33	Linezolid	=	2	1	4	1
33	Tetracycline	=	32	8	32	1
33	Vancomycin	=	2	1	4	1
34	Chloramphenicol	=	8	4	16	1
34	Ciprofloxacin	=	1	0,25	2	1
34	Daptomycin	=	2	1	4	1
34	Erythromycin	=	2	1	4	1
34	Gentamicin	<=	8	4	16	1
34	Linezolid	=	2	1	4	1
34	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
34	Tetracycline	=	32	8	32	1
34	Tigecycline	=	0,12	0,03	0,125	1
34	Vancomycin	=	4	1	4	1
34	Ampicillin	=	2	0,5	2	1
34	Teicoplanin	<=	0,5	0,25	1	1
36	Ampicillin	=	2	0,5	2	1
36	Chloramphenicol	=	8	4	16	1
36	Daptomycin	=	2	1	4	1
36	Gentamicin	=	16	4	16	1
36	Linezolid	=	2	1	4	1
36	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
36	Teicoplanin	<=	0,5	0,25	1	1
36	Tetracycline	=	32	8	32	1
36	Tigecycline	=	0,06	0,03	0,125	1
36	Erythromycin	=	2	1	4	1
36	Ciprofloxacin	=	1	0,25	2	1
36	Vancomycin	=	4	1	4	1
37	Chloramphenicol	=	4	4	16	1
37	Ciprofloxacin	=	0,5	0,25	2	1
37	Erythromycin	=	2	1	4	1
37	Gentamicin	<=	8	4	16	1
37	Linezolid	=	2	1	4	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
37	Teicoplanin	<=	0,5	0,25	1	1
37	Tetracycline	=	16	8	32	1
37	Vancomycin	=	4	1	4	1
37	Ampicillin	=	1	0,5	2	1
37	Tigecycline	=	0,125	0,03	0,125	1
38	Ampicillin	<=	0,5	0,5	2	1
38	Chloramphenicol	=	8	4	16	1
38	Ciprofloxacin	=	1	0,25	2	1
38	Erythromycin	=	2	1	4	1
38	Gentamicin	<=	8	4	16	1
38	Linezolid	=	2	1	4	1
38	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
38	Teicoplanin	<=	0,5	0,25	1	1
38	Tigecycline	=	0,12	0,03	0,125	1
38	Daptomycin	=	4	1	4	1
38	Vancomycin	=	4	1	4	1
38	Tetracycline	=	32	8	32	1
39	Ampicillin	=	0,5	0,5	2	1
39	Chloramphenicol	=	4	4	16	1
39	Erythromycin	=	2	1	4	1
39	Gentamicin	=	4	4	16	1
39	Linezolid	=	1	1	4	1
39	Tetracycline	=	16	8	32	1
39	Vancomycin	=	2	1	4	1
40	Chloramphenicol	=	8	4	16	1
40	Ciprofloxacin	=	0,25	0,25	2	1
40	Daptomycin	=	1	1	4	1
40	Erythromycin	=	2	1	4	1
40	Gentamicin	=	8	4	16	1
40	Linezolid	=	1	1	4	1
40	Quinopristin-dalfopristin (Synercid)	=	2	2	8	1
40	Tigecycline	=	0,06	0,03	0,125	1
40	Vancomycin	=	2	1	4	1
40	Tetracycline	=	8	8	32	1
40	Ampicillin	=	1	0,5	2	1
40	Teicoplanin	=	0,5	0,25	1	1
42	Ampicillin	=	2	0,5	2	1
42	Chloramphenicol	=	8	4	16	1
42	Ciprofloxacin	=	2	0,25	2	1
42	Erythromycin	=	4	1	4	1
42	Gentamicin	=	16	4	16	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
42	Linezolid	=	2	1	4	1
42	Teicoplanin	<=	0,5	0,25	1	1
42	Tigecycline	=	0,25	0,03	0,125	0
42	Vancomycin	=	4	1	4	1
42	Daptomycin	=	4	1	4	1
42	Tetracycline	=	32	8	32	1
42	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
45	Ampicillin	=	1	0,5	2	1
45	Chloramphenicol	=	8	4	16	1
45	Ciprofloxacin	=	1	0,25	2	1
45	Daptomycin	=	1	1	4	1
45	Erythromycin	<=	1	1	4	1
45	Linezolid	=	2	1	4	1
45	Quinopristin-dalfopristin (Synercid)	=	4	2	8	1
45	Teicoplanin	<=	0,5	0,25	1	1
45	Tigecycline	=	0,12	0,03	0,125	1
45	Vancomycin	=	4	1	4	1
45	Tetracycline	=	32	8	32	1
45	Gentamicin	<=	8	4	16	1
56	Ampicillin	=	1	0,5	2	1
56	Chloramphenicol	=	8	4	16	1
56	Ciprofloxacin	=	0,5	0,25	2	1
56	Daptomycin	=	2	1	4	1
56	Gentamicin	<=	8	4	16	1
56	Linezolid	=	2	1	4	1
56	Teicoplanin	<=	0,5	0,25	1	1
56	Tetracycline	=	16	8	32	1
56	Tigecycline	=	0,06	0,03	0,125	1
56	Vancomycin	<=	1	1	4	1
56	Quinopristin-dalfopristin (Synercid)	=	2	2	8	1
56	Erythromycin	<=	1	1	4	1
58	Ampicillin	=	1	0,5	2	1
58	Ciprofloxacin	=	1	0,25	2	1
58	Daptomycin	=	2	1	4	1
58	Erythromycin	<=	1	1	4	1
58	Gentamicin	=	16	4	16	1
58	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
58	Teicoplanin	<=	0,5	0,25	1	1
58	Tetracycline	=	16	8	32	1
58	Vancomycin	=	4	1	4	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
58	Linezolid	=	2	1	4	1
58	Chloramphenicol	=	8	4	16	1
58	Tigecycline	=	0,12	0,03	0,125	1
60	Ampicillin	=	1	0,5	2	1
60	Chloramphenicol	=	8	4	16	1
60	Ciprofloxacin	=	0,5	0,25	2	1
60	Erythromycin	=	2	1	4	1
60	Gentamicin	<=	8	4	16	1
60	Linezolid	=	2	1	4	1
60	Quinopristin-dalfopristin (Synercid)	=	8	2	8	1
60	Tetracycline	=	16	8	32	1
60	Tigecycline	=	0,06	0,03	0,125	1
60	Daptomycin	=	2	1	4	1
60	Vancomycin	=	4	1	4	1
60	Teicoplanin	<=	0,5	0,25	1	1

Appendix 6b- Test results from reference strain *S. aureus* ATCC 29213

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
2	Vancomycin	<=	1	0,5	2	1
2	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
2	Linezolid	=	2	1	4	1
2	Mupirocin	<=	0,5			
2	Clindamycin	<=	0,12	0,06	0,25	1
2	Chloramphenicol	=	8	2	16	1
2	Ciprofloxacin	<=	0,25	0,12	0,5	1
2	Erythromycin	<=	0,25	0,25	1	1
2	Cefoxitin	=	4	1	4	1
2	Gentamicin	<=	1	0,12	1	1
2	Sulfamethoxazole	<=	64	32	128	1
2	Tetracycline	<=	0,5	0,12	1	1
2	Tiamulin	<=	0,5			
2	Trimethoprim	<=	2	1	4	1
9	Vancomycin	<=	1	0,5	2	1
9	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
9	Linezolid	=	2	1	4	1
9	Clindamycin	<=	0,12	0,06	0,25	1
9	Chloramphenicol	<=	4	2	16	1
9	Ciprofloxacin	<=	0,25	0,12	0,5	1
9	Erythromycin	=	0,5	0,25	1	1
9	Cefoxitin	=	2	1	4	1
9	Gentamicin	<=	1	0,12	1	1
9	Sulfamethoxazole	<=	64	32	128	1
9	Tetracycline	<=	0,5	0,12	1	1
9	Tiamulin	<=	0,5			
9	Trimethoprim	<=	2	1	4	1
11	Clindamycin	<=	0,25	0,06	0,25	1
11	Chloramphenicol	=	4	2	16	1
11	Ciprofloxacin	=	0,25	0,12	0,5	1
11	Erythromycin	=	0,5	0,25	1	1
11	Cefoxitin	=	1	1	4	1
11	Gentamicin	<=	0,5	0,12	1	1
11	Tetracycline	<=	0,5	0,12	1	1
11	Trimethoprim	=	2	1	4	1
12	Clindamycin	<=	0,25	0,06	0,25	1
12	Chloramphenicol	=	4	2	16	1
12	Ciprofloxacin	=	0,25	0,12	0,5	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
12	Erythromycin	<=	0,25	0,25	1	1
12	Cefoxitin	=	2	1	4	1
12	Gentamicin	<=	0,5	0,12	1	1
12	Tetracycline	<=	0,5	0,12	1	1
12	Trimethoprim	=	1	1	4	1
17	Vancomycin	<=	1	0,5	2	1
17	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
17	Linezolid	=	4	1	4	1
17	Mupirocin	<=	0,5			
17	Clindamycin	<=	0,12	0,06	0,25	1
17	Chloramphenicol	=	16	2	16	1
17	Ciprofloxacin	<=	0,25	0,12	0,5	1
17	Erythromycin	=	0,5	0,25	1	1
17	Cefoxitin	=	4	1	4	1
17	Gentamicin	<=	1	0,12	1	1
17	Sulfamethoxazole	<=	64	32	128	1
17	Tetracycline	=	1	0,12	1	1
17	Tiamulin	=	1			
17	Trimethoprim	<=	2	1	4	1
19	Vancomycin	<=	1	0,5	2	1
19	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
19	Linezolid	=	2	1	4	1
19	Mupirocin	<=	0,5			
19	Clindamycin	<=	0,12	0,06	0,25	1
19	Chloramphenicol	=	8	2	16	1
19	Ciprofloxacin	<=	0,25	0,12	0,5	1
19	Erythromycin	=	0,5	0,25	1	1
19	Cefoxitin	=	2	1	4	1
19	Gentamicin	<=	1	0,12	1	1
19	Sulfamethoxazole	<=	64	32	128	1
19	Tetracycline	<=	0,5	0,12	1	1
19	Tiamulin	=	0,5			
19	Trimethoprim	<=	2	1	4	1
20	Vancomycin	<=	1	0,5	2	1
20	Quinopristin-dalfopristin (Synercid)	=	1	0,25	1	1
20	Linezolid	=	4	1	4	1
20	Mupirocin	<=	0,5			
20	Clindamycin	<=	0,12	0,06	0,25	1
20	Chloramphenicol	=	16	2	16	1
20	Ciprofloxacin	<=	0,25	0,12	0,5	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
20	Erythromycin	=	0,5	0,25	1	1
20	Cefoxitin	=	4	1	4	1
20	Gentamicin	<=	1	0,12	1	1
20	Sulfamethoxazole	=	128	32	128	1
20	Tetracycline	=	1	0,12	1	1
20	Tiamulin	=	1			
20	Trimethoprim	<=	2	1	4	1
21	Quinopristin-dalfopristin (Synercid)	=	1	0,25	1	1
21	Tetracycline	=	1	0,12	1	1
21	Vancomycin	<=	1	0,5	2	1
21	Linezolid	=	2	1	4	1
21	Clindamycin	<=	0,12	0,06	0,25	1
21	Chloramphenicol	=	8	2	16	1
21	Ciprofloxacin	<=	0,25	0,12	0,5	1
21	Erythromycin	=	0,5	0,25	1	1
21	Cefoxitin	=	4	1	4	1
21	Gentamicin	<=	1	0,12	1	1
21	Sulfamethoxazole	<=	64	32	128	1
21	Trimethoprim	<=	2	1	4	1
22	Gentamicin	<=	1	0,12	1	1
22	Vancomycin	<=	1	0,5	2	1
22	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
22	Linezolid	=	2	1	4	1
22	Clindamycin	<=	0,12	0,06	0,25	1
22	Chloramphenicol	=	8	2	16	1
22	Ciprofloxacin	=	0,5	0,12	0,5	1
22	Erythromycin	=	0,5	0,25	1	1
22	Cefoxitin	=	2	1	4	1
22	Sulfamethoxazole	<=	64	32	128	1
22	Tetracycline	<=	0,5	0,12	1	1
22	Trimethoprim	<=	2	1	4	1
23	Ciprofloxacin	<=	0,25	0,12	0,5	1
23	Vancomycin	<=	1	0,5	2	1
23	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
23	Linezolid	=	2	1	4	1
23	Clindamycin	<=	0,12	0,06	0,25	1
23	Chloramphenicol	=	8	2	16	1
23	Erythromycin	=	0,5	0,25	1	1
23	Cefoxitin	=	1	1	4	1
23	Gentamicin	<=	1	0,12	1	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
23	Sulfamethoxazole	<=	64	32	128	1
23	Tetracycline	<=	0,5	0,12	1	1
23	Trimethoprim	<=	2	1	4	1
25	Clindamycin	=	0,12	0,06	0,25	1
25	Erythromycin	=	0,25	0,25	1	1
25	Sulfamethoxazole-Trimethoprim	<=	0,12	0	0,5	1
25	Tetracycline	=	0,5	0,12	1	1
26	Vancomycin	<=	1	0,5	2	1
26	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
26	Linezolid	=	2	1	4	1
26	Mupirocin	<=	0,5			
26	Clindamycin	=	0,25	0,06	0,25	1
26	Chloramphenicol	=	8	2	16	1
26	Ciprofloxacin	=	0,5	0,12	0,5	1
26	Erythromycin	<=	0,25	0,25	1	1
26	Cefoxitin	=	4	1	4	1
26	Gentamicin	<=	1	0,12	1	1
26	Sulfamethoxazole	<=	64	32	128	1
26	Sulfamethoxazole-Trimethoprim	=	0,25	0	0,5	1
26	Tetracycline	<=	0,5	0,12	1	1
26	Tiamulin	<=	0,5			
26	Trimethoprim	<=	2	1	4	1
29	Vancomycin	<=	1	0,5	2	1
29	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
29	Linezolid	=	2	1	4	1
29	Clindamycin	<=	0,12	0,06	0,25	1
29	Chloramphenicol	=	8	2	16	1
29	Ciprofloxacin	<=	0,25	0,12	0,5	1
29	Erythromycin	=	0,25	0,25	1	1
29	Cefoxitin	=	4	1	4	1
29	Gentamicin	<=	1	0,12	1	1
29	Sulfamethoxazole	<=	64	32	128	1
29	Tetracycline	<=	0,5	0,12	1	1
29	Trimethoprim	<=	2	1	4	1
30	Vancomycin	<=	1	0,5	2	1
30	Trimethoprim	<=	2	1	4	1
30	Linezolid	=	2	1	4	1
30	Mupirocin	<=	0,5			
30	Clindamycin	<=	0,12	0,06	0,25	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
30	Chloramphenicol	=	8	2	16	1
30	Ciprofloxacin	<=	0,25	0,12	0,5	1
30	Erythromycin	<=	0,5	0,25	1	1
30	Cefoxitin	=	4	1	4	1
30	Gentamicin	<=	1	0,12	1	1
30	Sulfamethoxazole	<=	64	32	128	1
30	Tetracycline	<=	0,5	0,12	1	1
30	Tiamulin	<=	0,5			
30	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
31	Erythromycin	<=	0,5	0,25	1	1
31	Trimethoprim	<=	2	1	4	1
31	Quinopristin-dalfopristin (Synercid)	<=	1	0,25	1	1
31	Linezolid	<=	1	1	4	1
31	Mupirocin	<=	1			
31	Clindamycin	<=	0,25	0,06	0,25	1
31	Chloramphenicol	<=	16	2	16	1
31	Ciprofloxacin	<=	0,25	0,12	0,5	1
31	Cefoxitin	<=	4	1	4	1
31	Gentamicin	<=	2	0,12	1	1
31	Sulfamethoxazole	<=	128	32	128	1
31	Sulfamethoxazole-Trimethoprim	<=	0,5	0	0,5	1
31	Tetracycline	<=	1	0,12	1	1
31	Tiamulin	<=	2			
31	Vancomycin	<=	1	0,5	2	1
33	Gentamicin	<=	0,5	0,12	1	1
33	Trimethoprim	=	1	1	4	1
33	Clindamycin	<=	0,25	0,06	0,25	1
33	Chloramphenicol	=	8	2	16	1
33	Ciprofloxacin	=	0,25	0,12	0,5	1
33	Erythromycin	=	1	0,25	1	1
33	Cefoxitin	=	4	1	4	1
33	Tetracycline	<=	0,5	0,12	1	1
34	Sulfamethoxazole	=	128	32	128	1
34	Gentamicin	<=	1	0,12	1	1
34	Tetracycline	=	1	0,12	1	1
34	Trimethoprim	<=	2	1	4	1
34	Mupirocin	<=	0,5			
34	Tiamulin	=	1			
34	Vancomycin	<=	1	0,5	2	1
34	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
34	Linezolid	=	4	1	4	1
34	Clindamycin	<=	0,12	0,06	0,25	1
34	Chloramphenicol	=	16	2	16	1
34	Ciprofloxacin	<=	0,25	0,12	0,5	1
34	Erythromycin	=	0,5	0,25	1	1
34	Cefoxitin	=	4	1	4	1
36	Tetracycline	<=	0,5	0,12	1	1
36	Tiamulin	<=	0,5			
36	Trimethoprim	<=	2	1	4	1
36	Vancomycin	<=	1	0,5	2	1
36	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
36	Linezolid	=	2	1	4	1
36	Mupirocin	<=	0,5			
36	Clindamycin	<=	0,12	0,06	0,25	1
36	Chloramphenicol	=	8	2	16	1
36	Ciprofloxacin	<=	0,25	0,12	0,5	1
36	Erythromycin	<=	0,25	0,25	1	1
36	Cefoxitin	=	4	1	4	1
36	Gentamicin	<=	1	0,12	1	1
36	Sulfamethoxazole	<=	64	32	128	1
37	Vancomycin	<=	1	0,5	2	1
37	Chloramphenicol	=	8	2	16	1
37	Ciprofloxacin	=	0,25	0,12	0,5	1
37	Erythromycin	=	0,25	0,25	1	1
37	Cefoxitin	=	4	1	4	1
37	Gentamicin	=	0,25	0,12	1	1
37	Sulfamethoxazole	=	16	32	128	0
37	Tetracycline	=	0,5	0,12	1	1
37	Trimethoprim	=	1	1	4	1
37	Linezolid	=	2	1	4	1
39	Clindamycin	<=	0,25	0,06	0,25	1
39	Chloramphenicol	=	8	2	16	1
39	Ciprofloxacin	=	0,5	0,12	0,5	1
39	Erythromycin	=	1	0,25	1	1
39	Gentamicin	<=	0,5	0,12	1	1
39	Tetracycline	<=	0,5	0,12	1	1
39	Trimethoprim	=	2	1	4	1
40	Vancomycin	=	1	0,5	2	1
40	Gentamicin	=	1	0,12	1	1
40	Clindamycin	=	0,12	0,06	0,25	1
40	Chloramphenicol	=	4	2	16	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
40	Ciprofloxacin	=	0,25	0,12	0,5	1
40	Erythromycin	=	0,25	0,25	1	1
40	Cefoxitin	=	2	1	4	1
40	Sulfamethoxazole	=	64	32	128	1
40	Sulfamethoxazole-Trimethoprim	<=	0,5	0	0,5	1
40	Tetracycline	=	0,5	0,12	1	1
40	Tiamulin	=	0,5			
40	Trimethoprim	=	2	1	4	1
40	Quinopristin-dalfopristin (Synercid)	=	0,5	0,25	1	1
40	Linezolid	=	1	1	4	1
42	Ciprofloxacin	=	0,5	0,12	0,5	1
42	Tetracycline	<=	0,5	0,12	1	1
42	Cefoxitin	=	4	1	4	1
42	Gentamicin	<=	1	0,12	1	1
42	Sulfamethoxazole	<=	64	32	128	1
42	Tiamulin	=	1			
42	Trimethoprim	<=	2	1	4	1
42	Vancomycin	<=	1	0,5	2	1
42	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
42	Linezolid	=	4	1	4	1
42	Clindamycin	<=	0,12	0,06	0,25	1
42	Chloramphenicol	=	8	2	16	1
42	Mupirocin	<=	0,5			
42	Erythromycin	=	0,5	0,25	1	1
45	Vancomycin	<=	1	0,5	2	1
45	Trimethoprim	<=	2	1	4	1
45	Linezolid	=	2	1	4	1
45	Mupirocin	<=	0,5			
45	Clindamycin	<=	0,12	0,06	0,25	1
45	Chloramphenicol	=	8	2	16	1
45	Ciprofloxacin	=	0,5	0,12	0,5	1
45	Erythromycin	<=	0,25	0,25	1	1
45	Cefoxitin	=	4	1	4	1
45	Gentamicin	<=	1	0,12	1	1
45	Sulfamethoxazole	=	128	32	128	1
45	Tetracycline	=	1	0,12	1	1
45	Tiamulin	<=	0,5			
45	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
56	Vancomycin	<=	1	0,5	2	1

LAB	ANTIMICROBIAL	OPER	VALUE	MIN_VALUE	MAX_VALUE	SCORE
56	Ciprofloxacin	<=	0,25	0,12	0,5	1
56	Linezolid	=	2	1	4	1
56	Mupirocin	<=	0,5			
56	Clindamycin	<=	0,12	0,06	0,25	1
56	Chloramphenicol	=	8	2	16	1
56	Erythromycin	<=	0,25	0,25	1	1
56	Cefoxitin	=	1	1	4	1
56	Gentamicin	<=	1	0,12	1	1
56	Sulfamethoxazole	<=	64	32	128	1
56	Tetracycline	<=	0,5	0,12	1	1
56	Tiamulin	=	1			
56	Trimethoprim	<=	2	1	4	1
56	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1
58	Vancomycin	<=	1	0,5	2	1
58	Trimethoprim	<=	2	1	4	1
58	Linezolid	=	2	1	4	1
58	Mupirocin	<=	0,5			
58	Clindamycin	<=	0,12	0,06	0,25	1
58	Chloramphenicol	=	8	2	16	1
58	Ciprofloxacin	<=	0,25	0,12	0,5	1
58	Erythromycin	<=	0,25	0,25	1	1
58	Cefoxitin	=	4	1	4	1
58	Gentamicin	<=	1	0,12	1	1
58	Sulfamethoxazole	<=	64	32	128	1
58	Tetracycline	<=	0,5	0,12	1	1
58	Tiamulin	<=	0,5			
58	Quinopristin-dalfopristin (Synercid)	<=	0,5	0,25	1	1

Appendix 6c- Test results from reference strain *E. coli* ATCC 25922

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
2	Ampicillin	1	=	8	2	8	1
2	Azithromycin	1	=	4			
2	Cefotaxime	1	<=	0,25	0,03	0,12	1
2	Ceftazidime	1	<=	0,5	0,06	0,5	1
2	Chloramphenicol	1	<=	8	2	8	1
2	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
2	Colistin	1	<=	1	0,25	2	1
2	Gentamicin	1	<=	0,5	0,25	1	1
2	Meropenem	1	<=	0,03	0,008	0,06	1
2	Nalidixic acid	1	<=	4	1	4	1
2	Sulfamethoxazole	1	=	16	8	32	1
2	Tetracycline	1	<=	2	0,5	2	1
2	Tigecycline	1	<=	0,25	0,03	0,25	1
2	Trimethoprim	1	=	0,5	0,5	2	1
2	Cefepime	2	<=	0,06	0,015	0,12	1
2	Cefotaxime	2	<=	0,25	0,03	0,12	1
2	Cefotaxime/clavulanic acid	2	<=	0,06			
2	Cefoxitin	2	=	2	2	8	1
2	Ceftazidime	2	<=	0,25	0,06	0,5	1
2	Ceftazidime/clavulanic acid	2	<=	0,12			
2	Ertapenem	2	<=	0,015	0,004	0,015	1
2	Imipenem	2	<=	0,12	0,06	0,25	1
2	Meropenem	2	<=	0,03	0,008	0,06	1
2	Temocillin	2	=	8			
9	Ampicillin	1	=	4	2	8	1
9	Ceftazidime	1	<=	0,5	0,06	0,5	1
9	Chloramphenicol	1	<=	8	2	8	1
9	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
9	Colistin	1	<=	1	0,25	2	1
9	Gentamicin	1	<=	0,5	0,25	1	1
9	Meropenem	1	<=	0,03	0,008	0,06	1
9	Nalidixic acid	1	<=	4	1	4	1
9	Sulfamethoxazole	1	=	16	8	32	1
9	Tetracycline	1	<=	2	0,5	2	1
9	Tigecycline	1	<=	0,25	0,03	0,25	1
9	Trimethoprim	1	=	1	0,5	2	1
9	Cefepime	2	=	0,06	0,015	0,12	1
9	Cefotaxime	2	<=	0,25	0,03	0,12	1
9	Cefoxitin	2	=	4	2	8	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
9	Ceftazidime	2	<=	0,25	0,06	0,5	1
9	Ertapenem	2	<=	0,015	0,004	0,015	1
9	Imipenem	2	<=	0,12	0,06	0,25	1
9	Meropenem	2	<=	0,03	0,008	0,06	1
11	Ampicillin	1	=	2	2	8	1
11	Azithromycin	1	=	8			
11	Cefotaxime	1	<=	0,25	0,03	0,12	1
11	Ceftazidime	1	<=	0,5	0,06	0,5	1
11	Chloramphenicol	1	<=	8	2	8	1
11	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
11	Colistin	1	<=	1	0,25	2	1
11	Gentamicin	1	<=	0,5	0,25	1	1
11	Meropenem	1	<=	0,03	0,008	0,06	1
11	Nalidixic acid	1	<=	4	1	4	1
11	Sulfamethoxazole	1	=	16	8	32	1
11	Tetracycline	1	<=	2	0,5	2	1
11	Tigecycline	1	<=	0,25	0,03	0,25	1
11	Trimethoprim	1	=	0,5	0,5	2	1
11	Cefepime	2	<=	0,6	0,015	0,12	1
11	Cefotaxime	2	<=	0,25	0,03	0,12	1
11	Cefotaxime/clavulanic acid	2	<=	0,06			
11	Cefoxitin	2	=	2	2	8	1
11	Ceftazidime	2	<=	0,25	0,06	0,5	1
11	Ceftazidime/clavulanic acid	2	<=	0,12			
11	Ertapenem	2	<=	0,015	0,004	0,015	1
11	Imipenem	2	<=	0,12	0,06	0,25	1
11	Meropenem	2	<=	0,03	0,008	0,06	1
11	Temocillin	2	=	8			
12	Ampicillin	1	=	8	2	8	1
12	Azithromycin	1	=	8			
12	Cefotaxime	1	<=	0,25	0,03	0,12	1
12	Ceftazidime	1	<=	0,5	0,06	0,5	1
12	Chloramphenicol	1	<=	8	2	8	1
12	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
12	Colistin	1	<=	1	0,25	2	1
12	Gentamicin	1	<=	0,5	0,25	1	1
12	Meropenem	1	<=	0,03	0,008	0,06	1
12	Nalidixic acid	1	<=	4	1	4	1
12	Sulfamethoxazole	1	<=	8	8	32	1
12	Tetracycline	1	<=	2	0,5	2	1
12	Tigecycline	1	<=	0,25	0,03	0,25	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
12	Trimethoprim	1	=	1	0,5	2	1
12	Cefepime	2	<=	0,06	0,015	0,12	1
12	Cefotaxime	2	<=	0,25	0,03	0,12	1
12	Cefoxitin	2	=	2	2	8	1
12	Ceftazidime	2	<=	0,25	0,06	0,5	1
12	Ertapenem	2	<=	0,015	0,004	0,015	1
12	Imipenem	2	=	0,25	0,06	0,25	1
12	Meropenem	2	<=	0,03	0,008	0,06	1
12	Temocillin	2	=	16			
16	Ampicillin	1	=	4	2	8	1
16	Azithromycin	1	=	4			
16	Cefotaxime	1	<=	0,25	0,03	0,12	1
16	Ceftazidime	1	<=	0,5	0,06	0,5	1
16	Chloramphenicol	1	<=	8	2	8	1
16	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
16	Colistin	1	<=	1	0,25	2	1
16	Gentamicin	1	<=	0,5	0,25	1	1
16	Meropenem	1	<=	0,03	0,008	0,06	1
16	Nalidixic acid	1	<=	4	1	4	1
16	Sulfamethoxazole	1	=	64	8	32	0
16	Tetracycline	1	<=	2	0,5	2	1
16	Tigecycline	1	<=	0,25	0,03	0,25	1
16	Trimethoprim	1	=	1	0,5	2	1
16	Cefepime	2	<=	0,06	0,015	0,12	1
16	Cefotaxime	2	<=	0,25	0,03	0,12	1
16	Cefotaxime/clavulanic acid	2	<=	0,06			
16	Cefoxitin	2	=	4	2	8	1
16	Ceftazidime	2	<=	0,25	0,06	0,5	1
16	Ceftazidime/clavulanic acid	2	=	0,25			
16	Ertapenem	2	<=	0,015	0,004	0,015	1
16	Imipenem	2	=	0,25	0,06	0,25	1
16	Meropenem	2	<=	0,03	0,008	0,06	1
16	Temocillin	2	=	16			
17	Ampicillin	1	=	4	2	8	1
17	Azithromycin	1	=	8			
17	Cefotaxime	1	<=	0,12	0,03	0,12	1
17	Ceftazidime	1	<=	0,5	0,06	0,5	1
17	Chloramphenicol	1	<=	8	2	8	1
17	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
17	Colistin	1	<=	1	0,25	2	1
17	Gentamicin	1	<=	0,5	0,25	1	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
17	Meropenem	1	<=	0,03	0,008	0,06	1
17	Nalidixic acid	1	<=	4	1	4	1
17	Sulfamethoxazole	1	=	32	8	32	1
17	Tetracycline	1	<=	2	0,5	2	1
17	Tigecycline	1	<=	0,25	0,03	0,25	1
17	Trimethoprim	1	=	0,5	0,5	2	1
17	Cefepime	2	<=	0,06	0,015	0,12	1
17	Cefotaxime	2	<=	0,12	0,03	0,12	1
17	Cefotaxime/clavulanic acid	2	<=	0,06			
17	Cefoxitin	2	=	4	2	8	1
17	Ceftazidime	2	<=	0,25	0,06	0,5	1
17	Ceftazidime/clavulanic acid	2	<=	0,12			
17	Ertapenem	2	<=	0,015	0,004	0,015	1
17	Imipenem	2	<=	0,12	0,06	0,25	1
17	Meropenem	2	<=	0,03	0,008	0,06	1
17	Temocillin	2	=	8			
19	Ampicillin	1	=	4	2	8	1
19	Azithromycin	1	=	4			
19	Cefotaxime	1	<=	0,25	0,03	0,12	1
19	Ceftazidime	1	<=	0,5	0,06	0,5	1
19	Chloramphenicol	1	<=	8	2	8	1
19	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
19	Colistin	1	<=	1	0,25	2	1
19	Gentamicin	1	<=	0,5	0,25	1	1
19	Meropenem	1	<=	0,03	0,008	0,06	1
19	Nalidixic acid	1	=	8	1	4	0
19	Sulfamethoxazole	1	=	32	8	32	1
19	Tetracycline	1	<=	2	0,5	2	1
19	Tigecycline	1	<=	0,25	0,03	0,25	1
19	Trimethoprim	1	=	0,5	0,5	2	1
19	Cefepime	2	<=	0,06	0,015	0,12	1
19	Cefotaxime	2	<=	0,25	0,03	0,12	1
19	Cefotaxime/clavulanic acid	2	<=	0,06			
19	Cefoxitin	2	=	4	2	8	1
19	Ceftazidime	2	<=	0,25	0,06	0,5	1
19	Ceftazidime/clavulanic acid	2	<=	0,12			
19	Ertapenem	2	<=	0,015	0,004	0,015	1
19	Imipenem	2	<=	0,12	0,06	0,25	1
19	Meropenem	2	<=	0,03	0,008	0,06	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
19	Temocillin	2	=	8			
20	Ampicillin	1	=	4	2	8	1
20	Azithromycin	1	=	4			
20	Cefotaxime	1	<=	0,25	0,03	0,12	1
20	Ceftazidime	1	<=	0,5	0,06	0,5	1
20	Chloramphenicol	1	<=	8	2	8	1
20	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
20	Colistin	1	<=	1	0,25	2	1
20	Gentamicin	1	<=	0,5	0,25	1	1
20	Meropenem	1	<=	0,03	0,008	0,06	1
20	Nalidixic acid	1	<=	4	1	4	1
20	Sulfamethoxazole	1	=	32	8	32	1
20	Tetracycline	1	<=	2	0,5	2	1
20	Tigecycline	1	<=	0,25	0,03	0,25	1
20	Trimethoprim	1	=	0,5	0,5	2	1
20	Cefepime	2	<=	0,06	0,015	0,12	1
20	Cefotaxime	2	<=	0,25	0,03	0,12	1
20	Cefotaxime/clavulanic acid	2	<=	0,06			
20	Cefoxitin	2	=	4	2	8	1
20	Ceftazidime	2	<=	0,25	0,06	0,5	1
20	Ceftazidime/clavulanic acid	2	<=	0,12			
20	Ertapenem	2	<=	0,015	0,004	0,015	1
20	Imipenem	2	=	0,25	0,06	0,25	1
20	Meropenem	2	<=	0,03	0,008	0,06	1
20	Temocillin	2	=	16			
21	Ampicillin	1	=	4	2	8	1
21	Cefotaxime	1	<=	0,25	0,03	0,12	1
21	Ceftazidime	1	=	0,5	0,06	0,5	1
21	Chloramphenicol	1	=	8	2	8	1
21	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
21	Colistin	1	=	1	0,25	2	1
21	Gentamicin	1	=	0,5	0,25	1	1
21	Meropenem	1	=	0,03	0,008	0,06	1
21	Nalidixic acid	1	=	4	1	4	1
21	Sulfamethoxazole	1	=	32	8	32	1
21	Tetracycline	1	=	2	0,5	2	1
21	Tigecycline	1	=	0,25	0,03	0,25	1
21	Trimethoprim	1	=	1	0,5	2	1
21	Cefepime	2	=	0,06	0,015	0,12	1
21	Cefoxitin	2	=	2	2	8	1
21	Ertapenem	2	<=	0,015	0,004	0,015	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
21	Imipenem	2	<=	0,12	0,06	0,25	1
21	Meropenem	2	<=	0,03	0,008	0,06	1
22	Ampicillin	1	=	4	2	8	1
22	Azithromycin	1	=	4			
22	Cefotaxime	1	<=	0,25	0,03	0,12	1
22	Ceftazidime	1	<=	0,5	0,06	0,5	1
22	Chloramphenicol	1	<=	8	2	8	1
22	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
22	Colistin	1	<=	1	0,25	2	1
22	Gentamicin	1	=	1	0,25	1	1
22	Meropenem	1	<=	0,03	0,008	0,06	1
22	Nalidixic acid	1	<=	4	1	4	1
22	Sulfamethoxazole	1	=	16	8	32	1
22	Tetracycline	1	<=	2	0,5	2	1
22	Tigecycline	1	<=	0,25	0,03	0,25	1
22	Trimethoprim	1	=	2	0,5	2	1
23	Ampicillin	1	=	2	2	8	1
23	Azithromycin	1	<=	2			
23	Cefotaxime	1	<=	0,25	0,03	0,12	1
23	Ceftazidime	1	<=	0,5	0,06	0,5	1
23	Chloramphenicol	1	<=	8	2	8	1
23	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
23	Colistin	1	<=	1	0,25	2	1
23	Gentamicin	1	<=	0,5	0,25	1	1
23	Meropenem	1	<=	0,03	0,008	0,06	1
23	Nalidixic acid	1	<=	4	1	4	1
23	Sulfamethoxazole	1	=	16	8	32	1
23	Tetracycline	1	<=	2	0,5	2	1
23	Tigecycline	1	<=	0,25	0,03	0,25	1
23	Trimethoprim	1	=	0,5	0,5	2	1
23	Cefepime	2	<=	0,06	0,015	0,12	1
23	Cefotaxime	2	<=	0,25	0,03	0,12	1
23	Cefoxitin	2	=	2	2	8	1
23	Ceftazidime	2	<=	0,25	0,06	0,5	1
23	Ertapenem	2	<=	0,015	0,004	0,015	1
23	Imipenem	2	<=	0,12	0,06	0,25	1
23	Meropenem	2	<=	0,03	0,008	0,06	1
25	Ampicillin	1	=	8	2	8	1
25	Azithromycin	1	=	4			
25	Cefotaxime	1	<=	0,25	0,03	0,12	1
25	Ceftazidime	1	<=	0,5	0,06	0,5	1
25	Chloramphenicol	1	<=	8	2	8	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
25	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
25	Colistin	1	=	2	0,25	2	1
25	Gentamicin	1	<=	0,5	0,25	1	1
25	Meropenem	1	<=	0,03	0,008	0,06	1
25	Nalidixic acid	1	<=	4	1	4	1
25	Sulfamethoxazole	1	<=	8	8	32	1
25	Tetracycline	1	<=	2	0,5	2	1
25	Tigecycline	1	<=	0,25	0,03	0,25	1
25	Trimethoprim	1	=	0,5	0,5	2	1
25	Cefepime	2	<=	0,06	0,015	0,12	1
25	Cefotaxime	2	<=	0,25	0,03	0,12	1
25	Cefotaxime/clavulanic acid	2	<=	0,06			
25	Cefoxitin	2	=	4	2	8	1
25	Ceftazidime	2	<=	0,25	0,06	0,5	1
25	Ceftazidime/clavulanic acid	2	=	0,25			
25	Ertapenem	2	<=	0,015	0,004	0,015	1
25	Imipenem	2	=	0,25	0,06	0,25	1
25	Meropenem	2	<=	0,03	0,008	0,06	1
25	Temocillin	2	=	32			
26	Ampicillin	1	=	2	2	8	1
26	Azithromycin	1	<=	2			
26	Cefotaxime	1	<=	0,25	0,03	0,12	1
26	Ceftazidime	1	<=	0,5	0,06	0,5	1
26	Chloramphenicol	1	<=	8	2	8	1
26	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
26	Colistin	1	<=	1	0,25	2	1
26	Gentamicin	1	<=	0,5	0,25	1	1
26	Meropenem	1	<=	0,03	0,008	0,06	1
26	Nalidixic acid	1	<=	4	1	4	1
26	Sulfamethoxazole	1	<=	8	8	32	1
26	Tetracycline	1	<=	2	0,5	2	1
26	Tigecycline	1	<=	0,25	0,03	0,25	1
26	Trimethoprim	1	=	0,5	0,5	2	1
29	Ampicillin	1	=	2	2	8	1
29	Cefotaxime	1	<=	0,25	0,03	0,12	1
29	Ceftazidime	1	<=	0,5	0,06	0,5	1
29	Chloramphenicol	1	<=	8	2	8	1
29	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
29	Colistin	1	<=	1	0,25	2	1
29	Gentamicin	1	=	1	0,25	1	1
29	Meropenem	1	<=	0,03	0,008	0,06	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
29	Nalidixic acid	1	<=	4	1	4	1
29	Sulfamethoxazole	1	=	32	8	32	1
29	Tetracycline	1	<=	2	0,5	2	1
29	Tigecycline	1	<=	0,25	0,03	0,25	1
29	Trimethoprim	1	=	0,5	0,5	2	1
29	Cefepime	2	<=	0,06	0,015	0,12	1
29	Cefotaxime	2	<=	0,25	0,03	0,12	1
29	Cefoxitin	2	=	2	2	8	1
29	Ceftazidime	2	<=	0,25	0,06	0,5	1
29	Ertapenem	2	<=	0,015	0,004	0,015	1
29	Imipenem	2	<=	0,12	0,06	0,25	1
29	Meropenem	2	<=	0,03	0,008	0,06	1
30	Ampicillin	1	=	4	2	8	1
30	Azithromycin	1	=	4			
30	Cefotaxime	1	<=	0,25	0,03	0,12	1
30	Ceftazidime	1	<=	0,5	0,06	0,5	1
30	Chloramphenicol	1	<=	8	2	8	1
30	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
30	Colistin	1	<=	1	0,25	2	1
30	Gentamicin	1	<=	0,5	0,25	1	1
30	Meropenem	1	<=	0,03	0,008	0,06	1
30	Nalidixic acid	1	<=	4	1	4	1
30	Sulfamethoxazole	1	=	16	8	32	1
30	Tetracycline	1	<=	2	0,5	2	1
30	Tigecycline	1	<=	0,25	0,03	0,25	1
30	Trimethoprim	1	=	0,5	0,5	2	1
30	Cefepime	2	<=	0,06	0,015	0,12	1
30	Cefotaxime	2	<=	0,25	0,03	0,12	1
30	Cefotaxime/clavulanic acid	2	<=	0,06			
30	Cefoxitin	2	=	2	2	8	1
30	Ceftazidime	2	<=	0,25	0,06	0,5	1
30	Ceftazidime/clavulanic acid	2	=	0,25			
30	Ertapenem	2	<=	0,015	0,004	0,015	1
30	Imipenem	2	=	0,25	0,06	0,25	1
30	Meropenem	2	<=	0,03	0,008	0,06	1
30	Temocillin	2	=	16			
33	Ampicillin	1	=	4	2	8	1
33	Azithromycin	1	=	4			
33	Cefotaxime	1	<=	0,25	0,03	0,12	1
33	Ceftazidime	1	<=	0,5	0,06	0,5	1
33	Chloramphenicol	1	<=	8	2	8	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
33	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
33	Colistin	1	<=	1	0,25	2	1
33	Gentamicin	1	<=	0,5	0,25	1	1
33	Meropenem	1	<=	0,03	0,008	0,06	1
33	Nalidixic acid	1	<=	4	1	4	1
33	Sulfamethoxazole	1	=	32	8	32	1
33	Tetracycline	1	<=	2	0,5	2	1
33	Tigecycline	1	<=	0,25	0,03	0,25	1
33	Trimethoprim	1	=	0,5	0,5	2	1
33	Cefepime	2	<=	0,06	0,015	0,12	1
33	Cefotaxime	2	<=	0,25	0,03	0,12	1
33	Cefotaxime/clavulanic acid	2	<=	0,06			
33	Cefoxitin	2	=	4	2	8	1
33	Ceftazidime	2	<=	0,25	0,06	0,5	1
33	Ceftazidime/clavulanic acid	2	<=	0,12			
33	Ertapenem	2	<=	0,015	0,004	0,015	1
33	Imipenem	2	=	0,25	0,06	0,25	1
33	Meropenem	2	<=	0,03	0,008	0,06	1
33	Temocillin	2	=	8			
34	Ampicillin	1	=	4	2	8	1
34	Azithromycin	1	=	4			
34	Cefotaxime	1	<=	0,25	0,03	0,12	1
34	Ceftazidime	1	<=	0,5	0,06	0,5	1
34	Chloramphenicol	1	<=	8	2	8	1
34	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
34	Colistin	1	<=	1	0,25	2	1
34	Gentamicin	1	<=	0,5	0,25	1	1
34	Meropenem	1	=	0,06	0,008	0,06	1
34	Nalidixic acid	1	<=	4	1	4	1
34	Sulfamethoxazole	1	=	16	8	32	1
34	Tetracycline	1	<=	2	0,5	2	1
34	Tigecycline	1	<=	0,25	0,03	0,25	1
34	Trimethoprim	1	=	1	0,5	2	1
34	Cefepime	2	<=	0,06	0,015	0,12	1
34	Cefotaxime	2	<=	0,25	0,03	0,12	1
34	Cefotaxime/clavulanic acid	2	<=	0,06			
34	Cefoxitin	2	=	4	2	8	1
34	Ceftazidime	2	<=	0,25	0,06	0,5	1
34	Ceftazidime/clavulanic acid	2	<=	0,12			

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
34	Ertapenem	2	<=	0,015	0,004	0,015	1
34	Imipenem	2	<=	0,12	0,06	0,25	1
34	Meropenem	2	<=	0,03	0,008	0,06	1
34	Temocillin	2	=	8			
36	Ampicillin	1	=	8	2	8	1
36	Azithromycin	1	=	4			
36	Cefotaxime	1	<=	0,25	0,03	0,12	1
36	Ceftazidime	1	<=	0,5	0,06	0,5	1
36	Chloramphenicol	1	<=	8	2	8	1
36	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
36	Colistin	1	<=	1	0,25	2	1
36	Gentamicin	1	<=	0,5	0,25	1	1
36	Meropenem	1	<=	0,03	0,008	0,06	1
36	Nalidixic acid	1	<=	4	1	4	1
36	Sulfamethoxazole	1	=	16	8	32	1
36	Tetracycline	1	<=	2	0,5	2	1
36	Tigecycline	1	<=	0,25	0,03	0,25	1
36	Trimethoprim	1	=	0,5	0,5	2	1
36	Cefepime	2	<=	0,06	0,015	0,12	1
36	Cefotaxime	2	<=	0,25	0,03	0,12	1
36	Cefotaxime/clavulanic acid	2	<=	0,06			
36	Cefoxitin	2	=	4	2	8	1
36	Ceftazidime	2	<=	0,25	0,06	0,5	1
36	Ceftazidime/clavulanic acid	2	<=	0,25			
36	Ertapenem	2	<=	0,015	0,004	0,015	1
36	Imipenem	2	=	0,25	0,06	0,25	1
36	Meropenem	2	<=	0,03	0,008	0,06	1
36	Temocillin	2	=	16			
37	Ampicillin	1	=	4	2	8	1
37	Azithromycin	1	=	4			
37	Cefotaxime	1	<=	0,25	0,03	0,12	1
37	Ceftazidime	1	<=	0,5	0,06	0,5	1
37	Chloramphenicol	1	<=	8	2	8	1
37	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
37	Colistin	1	<=	1	0,25	2	1
37	Gentamicin	1	<=	0,5	0,25	1	1
37	Meropenem	1	<=	0,03	0,008	0,06	1
37	Nalidixic acid	1	<=	4	1	4	1
37	Sulfamethoxazole	1	=	32	8	32	1
37	Tetracycline	1	<=	2	0,5	2	1
37	Tigecycline	1	<=	0,25	0,03	0,25	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
37	Trimethoprim	1	=	1	0,5	2	1
38	Ampicillin	1	=	8	2	8	1
38	Azithromycin	1	=	4			
38	Cefotaxime	1	<=	0,25	0,03	0,12	1
38	Ceftazidime	1	<=	0,5	0,06	0,5	1
38	Chloramphenicol	1	<=	8	2	8	1
38	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
38	Colistin	1	<=	1	0,25	2	1
38	Gentamicin	1	<=	0,5	0,25	1	1
38	Meropenem	1	<=	0,03	0,008	0,06	1
38	Nalidixic acid	1	<=	4	1	4	1
38	Sulfamethoxazole	1	=	32	8	32	1
38	Tetracycline	1	<=	2	0,5	2	1
38	Tigecycline	1	<=	0,25	0,03	0,25	1
38	Trimethoprim	1	=	0,5	0,5	2	1
38	Cefepime	2	<=	0,06	0,015	0,12	1
38	Cefotaxime	2	<=	0,25	0,03	0,12	1
38	Cefotaxime/clavulanic acid	2	<=	0,06			
38	Cefoxitin	2	=	2	2	8	1
38	Ceftazidime	2	<=	0,25	0,06	0,5	1
38	Ceftazidime/clavulanic acid	2	<=	0,12			
38	Ertapenem	2	<=	0,015	0,004	0,015	1
38	Imipenem	2	=	0,25	0,06	0,25	1
38	Meropenem	2	<=	0,03	0,008	0,06	1
38	Temocillin	2	=	16			
39	Ampicillin	1	=	8	2	8	1
39	Azithromycin	1	=	4			
39	Cefotaxime	1	<=	0,25	0,03	0,12	1
39	Ceftazidime	1	<=	0,5	0,06	0,5	1
39	Chloramphenicol	1	<=	8	2	8	1
39	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
39	Colistin	1	<=	1	0,25	2	1
39	Gentamicin	1	<=	0,5	0,25	1	1
39	Meropenem	1	<=	0,03	0,008	0,06	1
39	Nalidixic acid	1	<=	4	1	4	1
39	Sulfamethoxazole	1	=	16	8	32	1
39	Tetracycline	1	<=	2	0,5	2	1
39	Tigecycline	1	<=	0,25	0,03	0,25	1
39	Trimethoprim	1	=	0,5	0,5	2	1
39	Cefepime	2	<=	0,06	0,015	0,12	1
39	Cefotaxime	2	<=	0,25	0,03	0,12	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
39	Cefoxitin	2	=	4	2	8	1
39	Ceftazidime	2	<=	0,25	0,06	0,5	1
39	Ertapenem	2	<=	0,015	0,004	0,015	1
39	Imipenem	2	=	0,25	0,06	0,25	1
39	Meropenem	2	<=	0,03	0,008	0,06	1
39	Temocillin	2	=	16			
40	Ampicillin	1	=	2	2	8	1
40	Cefotaxime	1	=	0,12	0,03	0,12	1
40	Ceftazidime	1	=	0,5	0,06	0,5	1
40	Chloramphenicol	1	=	8	2	8	1
40	Ciprofloxacin	1	=	0,015	0,004	0,015	1
40	Colistin	1	=	1	0,25	2	1
40	Gentamicin	1	=	1	0,25	1	1
40	Meropenem	1	=	0,06	0,008	0,06	1
40	Nalidixic acid	1	=	4	1	4	1
40	Sulfamethoxazole	1	=	32	8	32	1
40	Tetracycline	1	=	2	0,5	2	1
40	Tigecycline	1	=	0,25	0,03	0,25	1
40	Trimethoprim	1	=	0,5	0,5	2	1
40	Cefepime	2	=	0,12	0,015	0,12	1
40	Cefotaxime	2	=	0,12	0,03	0,12	1
40	Cefotaxime/clavulanic acid	2	=	0,12			
40	Cefoxitin	2	=	4	2	8	1
40	Ceftazidime	2	=	0,5	0,06	0,5	1
40	Ceftazidime/clavulanic acid	2	=	0,5			
40	Ertapenem	2	=	0,015	0,004	0,015	1
40	Imipenem	2	=	0,25	0,06	0,25	1
40	Meropenem	2	=	0,06	0,008	0,06	1
42	Ampicillin	1	=	4	2	8	1
42	Azithromycin	1	=	8			
42	Cefotaxime	1	<=	0,25	0,03	0,12	1
42	Ceftazidime	1	<=	0,5	0,06	0,5	1
42	Chloramphenicol	1	<=	8	2	8	1
42	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
42	Colistin	1	<=	1	0,25	2	1
42	Gentamicin	1	<=	0,5	0,25	1	1
42	Meropenem	1	<=	0,03	0,008	0,06	1
42	Nalidixic acid	1	<=	4	1	4	1
42	Sulfamethoxazole	1	=	32	8	32	1
42	Tetracycline	1	<=	2	0,5	2	1
42	Tigecycline	1	<=	0,25	0,03	0,25	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
42	Trimethoprim	1	=	0,5	0,5	2	1
42	Cefepime	2	<=	0,06	0,015	0,12	1
42	Cefotaxime	2	<=	0,25	0,03	0,12	1
42	Cefoxitin	2	=	4	2	8	1
42	Ceftazidime	2	<=	0,25	0,06	0,5	1
42	Ertapenem	2	<=	0,015	0,004	0,015	1
42	Imipenem	2	<=	0,12	0,06	0,25	1
42	Meropenem	2	<=	0,03	0,008	0,06	1
42	Temocillin	2	=	8			
45	Ampicillin	1	=	2	2	8	1
45	Azithromycin	1	<=	2			
45	Cefotaxime	1	<=	0,25	0,03	0,12	1
45	Ceftazidime	1	<=	0,5	0,06	0,5	1
45	Chloramphenicol	1	<=	8	2	8	1
45	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
45	Colistin	1	<=	1	0,25	2	1
45	Gentamicin	1	<=	0,5	0,25	1	1
45	Meropenem	1	<=	0,03	0,008	0,06	1
45	Nalidixic acid	1	<=	4	1	4	1
45	Sulfamethoxazole	1	=	16	8	32	1
45	Tetracycline	1	<=	2	0,5	2	1
45	Tigecycline	1	<=	0,25	0,03	0,25	1
45	Trimethoprim	1	=	0,5	0,5	2	1
45	Cefepime	2	<=	0,06	0,015	0,12	1
45	Cefotaxime	2	<=	0,25	0,03	0,12	1
45	Cefotaxime/clavulanic acid	2	<=	0,06			
45	Cefoxitin	2	=	4	2	8	1
45	Ceftazidime	2	<=	0,25	0,06	0,5	1
45	Ceftazidime/clavulanic acid	2	<=	0,12			
45	Ertapenem	2	<=	0,015	0,004	0,015	1
45	Imipenem	2	<=	0,12	0,06	0,25	1
45	Meropenem	2	<=	0,03	0,008	0,06	1
45	Temocillin	2	=	8			
56	Ampicillin	1	=	4	2	8	1
56	Azithromycin	1	=	4			
56	Cefotaxime	1	<=	0,25	0,03	0,12	1
56	Ceftazidime	1	<=	0,5	0,06	0,5	1
56	Chloramphenicol	1	<=	8	2	8	1
56	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
56	Colistin	1	<=	1	0,25	2	1
56	Gentamicin	1	<=	0,5	0,25	1	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
56	Meropenem	1	<=	0,03	0,008	0,06	1
56	Nalidixic acid	1	<=	4	1	4	1
56	Sulfamethoxazole	1	=	32	8	32	1
56	Tetracycline	1	<=	2	0,5	2	1
56	Tigecycline	1	<=	0,25	0,03	0,25	1
56	Trimethoprim	1	=	0,5	0,5	2	1
56	Cefepime	2	<=	0,06	0,015	0,12	1
56	Cefotaxime	2	<=	0,25	0,03	0,12	1
56	Cefotaxime/clavulanic acid	2	<=	0,06			
56	Cefoxitin	2	=	2	2	8	1
56	Ceftazidime	2	<=	0,25	0,06	0,5	1
56	Ceftazidime/clavulanic acid	2	<=	0,12			
56	Ertapenem	2	<=	0,015	0,004	0,015	1
56	Imipenem	2	<=	0,12	0,06	0,25	1
56	Meropenem	2	<=	0,03	0,008	0,06	1
56	Temocillin	2	=	8			
58	Ampicillin	1	=	8	2	8	1
58	Azithromycin	1	=	8			
58	Cefotaxime	1	<=	0,25	0,03	0,12	1
58	Ceftazidime	1	<=	0,5	0,06	0,5	1
58	Chloramphenicol	1	<=	8	2	8	1
58	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
58	Colistin	1	<=	1	0,25	2	1
58	Gentamicin	1	=	1	0,25	1	1
58	Meropenem	1	<=	0,03	0,008	0,06	1
58	Nalidixic acid	1	<=	4	1	4	1
58	Sulfamethoxazole	1	=	32	8	32	1
58	Tetracycline	1	<=	2	0,5	2	1
58	Tigecycline	1	<=	0,25	0,03	0,25	1
58	Trimethoprim	1	=	0,5	0,5	2	1
58	Cefepime	2	<=	0,06	0,015	0,12	1
58	Cefotaxime	2	<=	0,25	0,03	0,12	1
58	Cefotaxime/clavulanic acid	2	<=	0,06			
58	Cefoxitin	2	=	4	2	8	1
58	Ceftazidime	2	=	0,5	0,06	0,5	1
58	Ceftazidime/clavulanic acid	2	=	0,25			
58	Ertapenem	2	<=	0,015	0,004	0,015	1
58	Imipenem	2	<=	0,12	0,06	0,25	1
58	Meropenem	2	<=	0,03	0,008	0,06	1

LAB	ANTIMICROBIAL	PANEL	OPERATOR	VALUE	MIN_VALUE	MAX_VALUE	SCORE
58	Temocillin	2	=	32			
60	Ampicillin	1	=	4	2	8	1
60	Azithromycin	1	=	4			
60	Cefotaxime	1	<=	0,25	0,03	0,12	1
60	Ceftazidime	1	<=	0,5	0,06	0,5	1
60	Chloramphenicol	1	<=	8	2	8	1
60	Ciprofloxacin	1	<=	0,015	0,004	0,015	1
60	Colistin	1	<=	1	0,25	2	1
60	Gentamicin	1	<=	0,5	0,25	1	1
60	Meropenem	1	<=	0,03	0,008	0,06	1
60	Nalidixic acid	1	<=	4	1	4	1
60	Sulfamethoxazole	1	<=	8	8	32	1
60	Tetracycline	1	<=	2	0,5	2	1
60	Tigecycline	1	<=	0,25	0,03	0,25	1
60	Trimethoprim	1	<=	0,25	0,5	2	0

Appendix 7a- Summary of results Enterococci trial

AB	EURL ENT-9.1		EURL ENT-9.2		EURL ENT-9.3		EURL ENT-9.4		EURL ENT-9.5		EURL ENT-9.6		EURL ENT-9.7		EURL ENT-9.8	
	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct
AMP	26	25	26	25	26	25	26	26	24	20	26	26	26	26	26	26
CHL	27	27	27	26	26	26	27	27	27	27	27	27	26	26	27	27
CIP	24	24	24	24	24	24	24	24	24	24	24	24	24	24	24	24
DAP	22	22	22	22	22	22			22	22	22	22	21	21	21	21
ERY	27	26	27	27	27	27	27	27	27	27	27	27	27	26	27	27
GEN	26	26	27	27	26	26	27	27	27	27	27	27	27	27	27	27
LZD	27	27	27	27	27	27	27	27	27	27	27	27	27	27	27	27
SYN									21	20						
TEI	23	23	23	23	23	23	23	23	22	22	23	23	23	23	23	23
TET	27	27	27	27	27	27	27	26	26	26	27	27	27	27	27	27
TGC	22	19	22	20			22	20	22	21	22	19	22	18	22	20
VAN	27	27	27	27	27	27	27	27	27	27	27	27	27	27	27	27
Total	278	273	279	275	276	269	257	254	296	290	279	276	277	272	278	276
	dev	percent dev	dev	percent dev	dev	percent dev	dev	percent dev	dev	percent dev	dev	percent dev	dev	percent dev	dev	percent dev
AMP	1	3,8%	1	3,8%	1	3,8%	0	0,0%	4	16,7%	0	0,0%	0	0,0%	0	0,0%
CHL	0	0,0%	1	3,7%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%
CIP	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%
DAP	0	0,0%	0	0,0%	0	0,0%	0	NA	0	0,0%	0	0,0%	0	0,0%	0	0,0%
ERY	1	3,7%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	1	3,7%	0	0,0%
GEN	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%
LZD	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%
SYN			0	NA			0	NA	1	4,8%						
TEI	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%
TET	0	0,0%	0	0,0%	0	0,0%	1	3,7%	0	0,0%	0	0,0%	0	0,0%	0	0,0%
TGC	3	13,6%	2	9,1%	0	NA	2	9,1%	1	4,5%	3	13,6%	4	18,2%	2	9,1%
VAN	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%	0	0,0%

Combinations subtracted from report as they caused more than 25% deviation

Appendix 7b- Summary of results Staphylococci trial

AB	EURL ST-9.1		EURL ST-9.2		EURL ST-9.3		EURL ST-9.4		EURL ST-9.5		EURL ST-9.6		EURL ST-9.7		EURL ST-9.8	
	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct
FOX	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23
CHL	24	24	24	24	24	24	24	24	24	24	24	23	24	24	24	23
CIP	24	24	24	24	24	24			24	24	24	24	24	24	24	24
CLN	24	24	24	24	24	24	24	19	22	22	24	22	24	24	24	24
ERY	25	25	24	22	24	24	25	21	25	25	25	25	25	25	25	25
GEN	24	24	24	24	24	24	24	23	24	23	24	24	24	24	24	24
LZD	21	21	21	21	21	21	20	20	21	21	21	20	21	21	21	21
MUP	19	19	19	19	19	19	19	19	19	19	19	19	19	19	19	19
SYN	19	19	18	17	17	15	18	16			18	18	18	18	18	18
SMX	20	19	20	20	20	20	19	18	20	20	20	20	20	20	20	20
SXT	6	6	5	5	5	5	5	5	6	6	6	6	6	6	5	5
TET	25	25	25	25	25	25	25	25	25	23	24	24	25	25	25	25
TIA	18	18	17	17	19	19	17	15	18	18	18	17	18	18	18	18
TMP	24	23	24	24	24	24	24	23	24	23	24	24	24	22	24	24
VAN	20	20	21	21	21	21	21	21	20	20	20	20	20	20	20	20
Total	316	314	313	310	314	312	288	272	295	291	314	309	315	313	314	313
	dev	per dev	dev	per dev	dev	per dev	dev	per dev	dev	per dev	dev	per dev	dev	per dev	dev	per dev
FOX	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%
CHL	0	0%	0	0%	0	0%	0	0%	0	0%	1	4%	0	0%	1	4%
CIP	0	0%	0	0%	0	0%	0	NA	0	0%	0	0%	0	0%	0	0%
CLN	0	0%	0	0%	0	0%	5	21%	0	0%	2	8%	0	0%	0	0%
ERY	0	0%	2	8%	0	0%	4	16%	0	0%	0	0%	0	0%	0	0%
GEN	0	0%	0	0%	0	0%	1	4%	1	4%	0	0%	0	0%	0	0%
LZD	0	0%	0	0%	0	0%	0	0%	0	0%	1	5%	0	0%	0	0%
MUP	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%
SYN	0	0%	1	6%	2	12%	2	11%	0	NA	0	0%	0	0%	0	0%
SMX	1	5%	0	0%	0	0%	1	5%	0	0%	0	0%	0	0%	0	0%
SXT	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%
TET	0	0%	0	0%	0	0%	0	0%	2	8%	0	0%	0	0%	0	0%
TIA	0	0%	0	0%	0	0%	2	12%	0	0%	1	6%	0	0%	0	0%
TMP	1	4%	0	0%	0	0%	1	4%	1	4%	0	0%	2	8%	0	0%
VAN	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%

Combinations subtracted from report as they caused more than 25% deviation.

Appendix 7c- Summary of results *E. coli* trial

	EURL EC-9.1		EURL EC-9.2		EURL EC-9.3		EURL EC-9.4		EURL EC-9.5		EURL EC-9.6		EURL EC-9.7		EURL EC-9.8	
	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct	tested	correct
AMP	31	31	31	31	31	30	31	30	31	31	30	29	31	31	31	31
AZI	29	28	29	29	28	28	28	28	28	28	28	28	29	29	28	28
FEP	23	18			30	30	30	27	31	30			31	31	31	31
FOT	54	48	31	31	61	60	61	61	62	61	30	29	62	62	62	62
FOX	23	22			30	30	31	31	30	30			31	31	31	30
TAZ	54	54	31	31	61	60	62	62	62	62	30	28	62	62	62	62
CHL	30	30	31	30	31	31	31	31	31	31	30	29	31	31	29	27
CIP	31	31	31	31	31	31	31	31	31	31	30	29	31	31	31	31
COL	31	31	31	31	31	31	31	31	31	30	30	30	31	31	31	31
ETP	23	23			30	29	31	31	31	31			31	31	31	31
GEN	31	31	31	31	31	31	31	31	31	31	30	29	31	31	31	31
IMI	22	20			30	30	31	31	31	31			31	31	31	31
MERO	54	36	31	31	61	61	62	61	61	61	30	30	62	62	62	62
NAL	31	31	31	31	31	31	31	31	31	29	30	30	31	31	31	31
SMX	31	31	31	30	31	31	31	31	31	31	30	27	31	31	31	31
TET	31	31	31	31	31	31	31	31	31	31	30	29	31	31	31	31
TGC	31	31	31	31	31	31	31	31	31	31	30	30	31	31	31	31
TMP	31	30	30	30	31	31	31	31	31	31	30	30	31	31	31	30
Total	591	557	431	429	641	637	646	641	646	641	418	407	649	649	646	642
AB	dev	per dev	dev	per dev	dev	per dev	dev	per dev	dev	per dev	dev	per dev	dev	per dev	dev	per dev
AMP	0	0%	0	0%	1	3%	1	3%	0	0%	1	3%	0	0%	0	0%
AZI	1	3%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%
FEP	5	22%	0	0%	0	0%	3	10%	1	3%	0	0%	0	0%	0	0%
FOT	6	11%	0	0%	1	2%	0	0%	1	2%	1	3%	0	0%	0	0%
FOX	1	4%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	1	3%
TAZ	0	0%	0	0%	1	2%	0	0%	0	0%	2	7%	0	0%	0	0%
CHL	0	0%	1	3%	0	0%	0	0%	0	0%	1	3%	0	0%	2	7%
CIP	0	0%	0	0%	0	0%	0	0%	0	0%	1	3%	0	0%	0	0%
COL	0	0%	0	0%	0	0%	0	0%	1	3%	0	0%	0	0%	0	0%
ETP	0	0%	0	0%	1	3%	0	0%	0	0%	0	0%	0	0%	0	0%
GEN	0	0%	0	0%	0	0%	0	0%	0	0%	1	3%	0	0%	0	0%
IMI	2	9%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%
MERO	18	33,3%	0	0%	0	0%	1	2%	0	0%	0	0%	0	0%	0	0%
NAL	0	0%	0	0%	0	0%	0	0%	2	6%	0	0%	0	0%	0	0%

SMX	0	0%	1	3%	0	0%	0	0%	0	0%	3	10%	0	0%	0	0%
TET	0	0%	0	0%	0	0%	0	0%	0	0%	1	3%	0	0%	0	0%
TGC	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%
TMP	1	3%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	1	3%



Combination subtracted from report as it caused more than 25% deviations.

Appendix 8a- Deviations of results Enterococci trial

LAB	STRAINID	ANTIMICROBIAL	EXP_INTERP	INTERP	READVALUE	EXPVAL
2	EURL ENT-9.2	Tigecycline TGC	S	R	0,5	0,25
2	EURL ENT-9.3	Tigecycline TGC	S	R	0,5	0,25
2	EURL ENT-9.4	Daptomycin DAP	R	S	4	8
2	EURL ENT-9.7	Tigecycline TGC	S	R	0,5	0,25
9	EURL ENT-9.4	Daptomycin DAP	R	S	4	8
11	EURL ENT-9.4	Daptomycin DAP	R	S	4	8
12	EURL ENT-9.1	Ampicillin AMP	S	R	16	1
12	EURL ENT-9.2	Ampicillin AMP	S	R	32	1
12	EURL ENT-9.3	Ampicillin AMP	S	R	16	1
12	EURL ENT-9.5	Ampicillin AMP	S	R	8	4
16	EURL ENT-9.4	Daptomycin DAP	R	S	4	8
16	EURL ENT-9.5	Ampicillin AMP	S	R	8	4
17	EURL ENT-9.4	Daptomycin DAP	R	S	4	8
19	EURL ENT-9.1	Tigecycline TGC	S	R	0,5	0,25
19	EURL ENT-9.3	Tigecycline TGC	S	R	0,5	0,25
19	EURL ENT-9.4	Daptomycin DAP	R	S	2	8
19	EURL ENT-9.4	Tigecycline TGC	S	R	0,5	0,25
19	EURL ENT-9.6	Tigecycline TGC	S	R	0,5	0,25
19	EURL ENT-9.7	Tigecycline TGC	S	R	0,5	0,25
19	EURL ENT-9.8	Tigecycline TGC	S	R	0,5	0,25
20	EURL ENT-9.4	Quinopristin-dalfopristin (Synercid) SYN	S	R	8	4
20	EURL ENT-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	4	4
22	EURL ENT-9.4	Daptomycin DAP	R	S	2	8
23	EURL ENT-9.4	Daptomycin DAP	R	S	2	8
25	EURL ENT-9.4	Daptomycin DAP	R	S	2	8
25	EURL ENT-9.5	Ampicillin AMP	S	R	8	4
26	EURL ENT-9.4	Daptomycin DAP	R	S	2	8
26	EURL ENT-9.4	Quinopristin-dalfopristin (Synercid) SYN	S	R	8	4
29	EURL ENT-9.3	Tigecycline TGC	S	R	0,5	0,25
29	EURL ENT-9.4	Daptomycin DAP	R	S	2	8
29	EURL ENT-9.7	Tigecycline TGC	S	R	0,5	0,25
30	EURL ENT-9.4	Daptomycin DAP	R	S	2	8
34	EURL ENT-9.1	Erythromycin ERY	R	S	<=1	> 128
34	EURL ENT-9.4	Daptomycin DAP	R	S	2	8
34	EURL ENT-9.4	Quinopristin-dalfopristin (Synercid) SYN	S	R	8	4
34	EURL ENT-9.7	Erythromycin ERY	R	S	<=1	> 128
36	EURL ENT-9.4	Daptomycin DAP	R	S	4	8
38	EURL ENT-9.1	Tigecycline TGC	S	R	0,5	0,25
38	EURL ENT-9.3	Tigecycline TGC	S	R	0,5	0,25


LAB	STRAINID	ANTIMICROBIAL	EXP_INTERP	INTERP	READVALUE	EXPVAL
38	EURL ENT-9.6	Tigecycline TGC	S	R	0,5	0,25
40	EURL ENT-9.2	Chloramphenicol CHL	R	S	<4	128
40	EURL ENT-9.4	Daptomycin DAP	R	S	1	8
40	EURL ENT-9.4	Tetracycline TET	R	S	2	128
42	EURL ENT-9.1	Tigecycline TGC	S	R	1	0,25
42	EURL ENT-9.2	Tigecycline TGC	S	R	0,5	0,25
42	EURL ENT-9.3	Tigecycline TGC	S	R	0,5	0,25
42	EURL ENT-9.4	Daptomycin DAP	R	S	4	8
42	EURL ENT-9.4	Quinopristin-dalfopristin (Synercid) SYN	S	R	8	4
42	EURL ENT-9.4	Tigecycline TGC	S	R	0,5	0,25
42	EURL ENT-9.5	Ampicillin AMP	S	R	8	4
42	EURL ENT-9.5	Tigecycline TGC	S	R	1	0,12
42	EURL ENT-9.6	Tigecycline TGC	S	R	0,5	0,25
42	EURL ENT-9.7	Tigecycline TGC	S	R	0,5	0,25
42	EURL ENT-9.8	Tigecycline TGC	S	R	0,5	0,25
45	EURL ENT-9.3	Tigecycline TGC	S	R	0,5	0,25
45	EURL ENT-9.4	Daptomycin DAP	R	S	2	8
56	EURL ENT-9.4	Daptomycin DAP	R	S	4	8
56	EURL ENT-9.4	Quinopristin-dalfopristin (Synercid) SYN	S	R	8	4
58	EURL ENT-9.4	Daptomycin DAP	R	S	4	8
58	EURL ENT-9.4	Quinopristin-dalfopristin (Synercid) SYN	S	R	8	4
60	EURL ENT-9.4	Daptomycin DAP	R	S	4	8
60	EURL ENT-9.4	Quinopristin-dalfopristin (Synercid) SYN	S	R	8	4

Combinations subtracted from report as they caused more than 25% deviation.

Appendix 8b- Deviations of results Staphylococci trial

LAB	STRAINID	ANTIMICROBIAL	EXP_INTERP	INTERP	READVALUE	EXPVAL
2	EURL ST-9.4	Ciprofloxacin CIP	R	S	1	2
2	EURL ST-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	2	1
11	EURL ST-9.1	Trimethoprim TMP	S	R	>32	<= 0.5
11	EURL ST-9.4	Ciprofloxacin CIP	R	S	1	2
11	EURL ST-9.4	Clindamycin CLN	S	R	>32	0,12
11	EURL ST-9.4	Erythromycin ERY	S	R	>32	0,25
12	EURL ST-9.4	Ciprofloxacin CIP	R	S	0,5	2
17	EURL ST-9.5	Gentamicin GEN	S	R	4	0,5
17	EURL ST-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	2	1
17	EURL ST-9.6	Chloramphenicol CHL	S	R	32	8
17	EURL ST-9.6	Linezolid LZD	S	R	8	4
19	EURL ST-9.2	Erythromycin ERY	S	R	>8	0,25
19	EURL ST-9.4	Ciprofloxacin CIP	R	S	1	2
20	EURL ST-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	2	1
20	EURL ST-9.5	Tetracycline TET	S	R	2	0,5
21	EURL ST-9.4	Ciprofloxacin CIP	R	S	1	2
22	EURL ST-9.4	Ciprofloxacin CIP	R	S	1	2
22	EURL ST-9.4	Clindamycin CLN	S	R	>4	0,12
22	EURL ST-9.4	Erythromycin ERY	S	R	>8	0,25
22	EURL ST-9.4	Quinopristin-dalfopristin (Synercid) SYN	S	R	>4	<= 0.5
22	EURL ST-9.4	Tiamulin TIA	S	R	>4	0,5
22	EURL ST-9.4	Trimethoprim TMP	S	R	4	1
22	EURL ST-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	2	1
23	EURL ST-9.3	Quinopristin-dalfopristin (Synercid) SYN	R	S	1	2
23	EURL ST-9.4	Ciprofloxacin CIP	R	S	1	2
23	EURL ST-9.7	Trimethoprim TMP	S	R	4	1
23	EURL ST-9.8	Chloramphenicol CHL	S	R	8	16
26	EURL ST-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	4	1
29	EURL ST-9.4	Ciprofloxacin CIP	R	S	<0.25	2
29	EURL ST-9.4	Clindamycin CLN	S	R	>4	0,12
29	EURL ST-9.4	Erythromycin ERY	S	R	>8	0,25
29	EURL ST-9.4	Gentamicin GEN	R	S	<1	> 16
29	EURL ST-9.4	Quinopristin-dalfopristin (Synercid) SYN	S	R	>4	<= 0.5
29	EURL ST-9.4	Sulfamethoxazole SMX	R	S	<64	256
29	EURL ST-9.4	Tiamulin TIA	S	R	>4	0,5
29	EURL ST-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	2	1
29	EURL ST-9.5	Tetracycline TET	S	R	>16	0,5
29	EURL ST-9.5	Trimethoprim TMP	S	R	>32	1

LAB	STRAINID	ANTIMICROBIAL	EXP_INTERP	INTERP	READVALUE	EXPVAL
29	EURL ST-9.6	Clindamycin CLN	S	R	0,5	0,25
29	EURL ST-9.6	Tiamulin TIA	S	R	>4	1
30	EURL ST-9.4	Ciprofloxacin CIP	R	S	1	2
31	EURL ST-9.2	Quinopristin-dalfopristin (Synercid) SYN	R	S	<=1	2
31	EURL ST-9.3	Quinopristin-dalfopristin (Synercid) SYN	R	S	<=1	2
33	EURL ST-9.4	Ciprofloxacin CIP	R	S	0,5	2
34	EURL ST-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	2	1
36	EURL ST-9.4	Ciprofloxacin CIP	R	S	1	2
37	EURL ST-9.4	Ciprofloxacin CIP	R	S	1	2
39	EURL ST-9.4	Ciprofloxacin CIP	R	S	1	2
39	EURL ST-9.4	Clindamycin CLN	S	R	>32	0,12
39	EURL ST-9.6	Clindamycin CLN	S	R	1	0,25
40	EURL ST-9.4	Ciprofloxacin CIP	R	S	<0.25	2
40	EURL ST-9.4	Clindamycin CLN	S	R	>4	0,12
40	EURL ST-9.4	Erythromycin ERY	S	R	>8	0,25
42	EURL ST-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	2	1
45	EURL ST-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	2	1
56	EURL ST-9.1	Sulfamethoxazole SMX	S	R	>512	<= 32
56	EURL ST-9.2	Erythromycin ERY	S	R	4	0,25
56	EURL ST-9.7	Trimethoprim TMP	S	R	>32	1
58	EURL ST-9.5	Quinopristin-dalfopristin (Synercid) SYN	S	R	2	1

 Combinations that were subtracted from report as they caused more than 25% deviation.

Appendix 8c- Deviations of results *E. coli* trial

LAB	STRAINID	PANEL	ANTIBIOTIC	READVALUE	EXPVAL	INTERP	EXP_INTERP
2	EURL EC-9.1	1	Meropenem MERO	0,12	0,5	S	R
2	EURL EC-9.1	2	Imipenem IMI	0,25	1	S	R
2	EURL EC-9.1	2	Meropenem MERO	0,12	0,5	S	R
2	EURL EC-9.6	1	Ciprofloxacin CIP	0,25	0,5	S	R
4	EURL EC-9.1	1	Meropenem MERO	0,12	0,5	S	R
4	EURL EC-9.1	2	Meropenem MERO	0,12	0,5	S	R
6	EURL EC-9.1	1	Cefotaxime FOT	0,5	<= 0.25	R	S
6	EURL EC-9.1	2	Cefotaxime FOT	0,5	<= 0.25	R	S
11	EURL EC-9.1	2	Cefepime FEP	0,25	0,12	R	S
11	EURL EC-9.1	2	Cefoxitin FOX	4	4	R	S
12	EURL EC-9.1	1	Cefotaxime FOT	0,5	<= 0.25	R	S
12	EURL EC-9.1	2	Cefepime FEP	0,25	0,12	R	S
17	EURL EC-9.1	1	Meropenem MERO	0,12	0,5	S	R
17	EURL EC-9.5	1	Nalidixic acid NAL	128	128	S	R
17	EURL EC-9.6	1	Gentamicin GEN	<=0.5	1	R	S
19	EURL EC-9.1	1	Meropenem MERO	0,12	0,5	S	R
19	EURL EC-9.8	2	Cefoxitin FOX	16	4	R	S
20	EURL EC-9.1	1	Meropenem MERO	0,12	0,5	S	R
20	EURL EC-9.1	2	Imipenem IMI	0,5	1	S	R
20	EURL EC-9.1	2	Meropenem MERO	0,12	0,5	S	R
21	EURL EC-9.1	1	Meropenem MERO	0,12	0,5	S	R
21	EURL EC-9.5	1	Nalidixic acid NAL	8	128	S	R
23	EURL EC-9.1	1	Meropenem MERO	0,12	0,5	S	R
23	EURL EC-9.4	1	Ampicillin AMP	>64	> 64	S	R
23	EURL EC-9.4	2	Cefepime FEP	0,12	0,25	S	R
25	EURL EC-9.1	2	Cefepime FEP	0,25	0,12	R	S
25	EURL EC-9.1	2	Cefotaxime FOT	0,5	<= 0.25	R	S
25	EURL EC-9.5	1	Colistin COL	<= 1	<= 1	R	S
26	EURL EC-9.1	1	Meropenem MERO	0,06	0,5	S	R
26	EURL EC-9.8	1	Chloramphenicol CHL	16	32	S	R
29	EURL EC-9.1	1	Meropenem MERO	0,12	0,5	S	R
30	EURL EC-9.4	2	Cefepime FEP	0,125	0,25	S	R
33	EURL EC-9.1	1	Meropenem MERO	0,06	0,5	S	R
33	EURL EC-9.6	1	Ampicillin AMP	>64	4	R	S
33	EURL EC-9.6	1	Cefotaxime FOT	4	<= 0.06	R	S
33	EURL EC-9.6	1	Ceftazidime TAZ	8	0,25	R	S
33	EURL EC-9.6	1	Chloramphenicol CHL	128	0,5	R	S
33	EURL EC-9.6	1	Sulfamethoxazole SMX	>1024	16	R	S
33	EURL EC-9.6	1	Tetracycline TET	64	4	R	S
34	EURL EC-9.4	2	Cefepime FEP	0,25	0,25	S	R
34	EURL EC-9.5	2	Cefepime FEP	0,25	0,25	S	R

LAB	STRAINID	PANEL	ANTIBIOTIC	READVALUE	EXPVAL	INTERP	EXP_INTERP
36	EURL EC-9.1	2	Cefepime FEP	0,25	0,12	R	S
39	EURL EC-9.1	1	Azithromycin AZI	<=2	> 64	S	R
39	EURL EC-9.1	1	Meropenem MERO	0,06	0,5	S	R
39	EURL EC-9.3	2	Ertapenem ETP	<=0.15	<= 0.15	R	S
39	EURL EC-9.4	1	Meropenem MERO	<=0.03	<= 0.03	R	S
39	EURL EC-9.6	1	Ceftazidime TAZ	<=4	0,25	R	S
40	EURL EC-9.1	1	Meropenem MERO	<0.03	0,5	S	R
40	EURL EC-9.1	1	Trimethoprim TMP	<0.25	> 32	S	R
40	EURL EC-9.6	1	Sulfamethoxazole SMX	1024	16	R	S
40	EURL EC-9.8	1	Chloramphenicol CHL	16	32	S	R
40	EURL EC-9.8	1	Trimethoprim TMP	<0.25	0,5	R	S
42	EURL EC-9.1	1	Meropenem MERO	0,06	0,5	S	R
45	EURL EC-9.1	1	Cefotaxime FOT	0,5	<= 0.25	R	S
45	EURL EC-9.1	2	Cefepime FEP	0,25	0,12	R	S
45	EURL EC-9.1	2	Cefotaxime FOT	0,5	<= 0.25	R	S
56	EURL EC-9.1	1	Meropenem MERO	0,12	0,5	S	R
56	EURL EC-9.1	2	Meropenem MERO	0,12	0,5	S	R
56	EURL EC-9.3	1	Ampicillin AMP	2	> 64	S	R
56	EURL EC-9.3	1	Cefotaxime FOT	<=0.25	64	S	R
56	EURL EC-9.3	1	Ceftazidime TAZ	<=0.5	2	S	R
59	EURL EC-9.2	1	Chloramphenicol CHL	16	8	R	S
59	EURL EC-9.2	1	Sulfamethoxazole SMX	1024	<= 8	R	S
59	EURL EC-9.5	1	Cefotaxime FOT	1	2	S	R
59	EURL EC-9.6	1	Sulfamethoxazole SMX	1024	16	R	S

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ISBN: 978-87-93109-85-8